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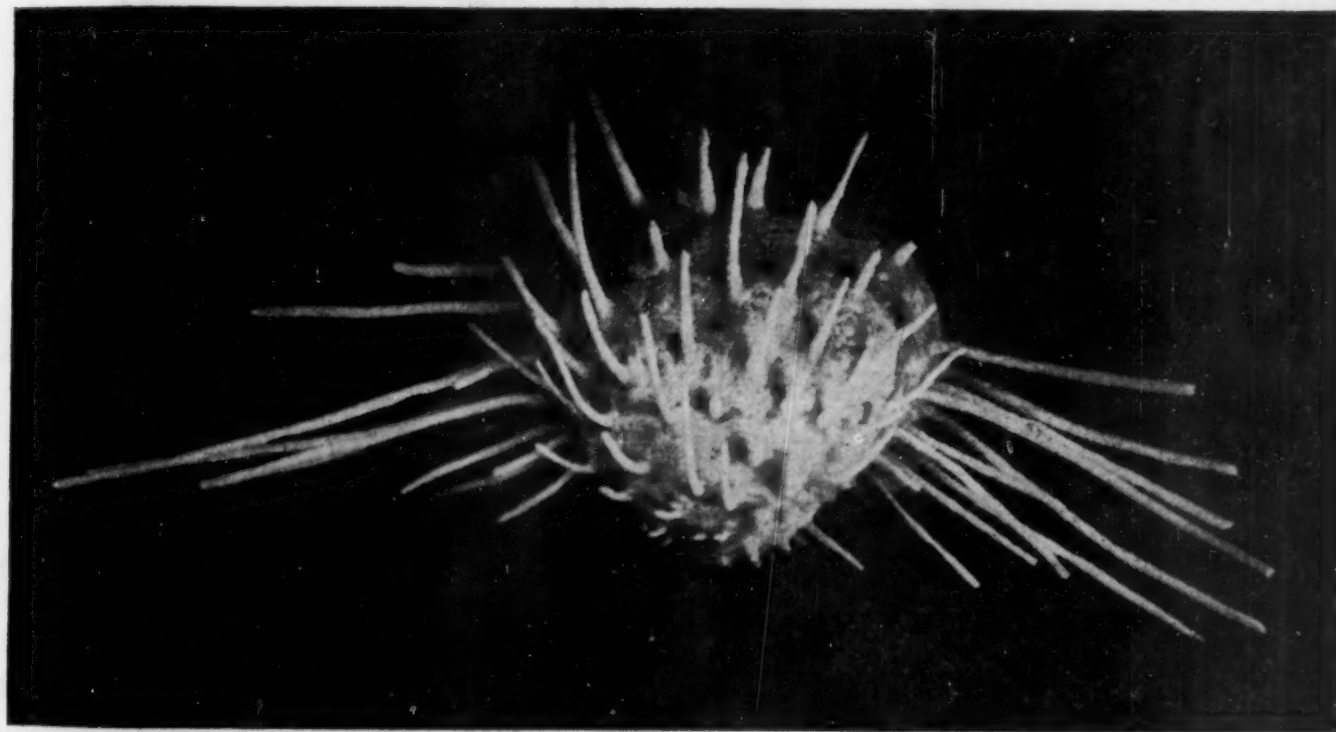
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Role of Microorganisms in Acid Mine Drainage  
Arthur R. Colmer and M. E. Hinkle

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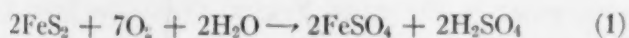
# The Role of Microorganisms in Acid Mine Drainage: A Preliminary Report

Arthur R. Colmer and M. E. Hinkle

*Agricultural and Engineering Experiment Stations, West Virginia University*

SOME WATERS ISSUING FROM BITUMINOUS coal mines are near neutral or alkaline in reaction, and, as such, are not considered to be of great importance in the problem of stream pollution. This is not the case with acid mine waters. Waters of this class are objectionable, and their disposal has engaged the attention of the many agencies concerned with water conservation and with the abatement of stream pollution. Carpenter and Herndon (1) and Hodge (2) have reported analyses of this type of mine drainage. Analyses of these waters vary widely from mine to mine and from coal seam to coal seam. One sample of acid mine water had a pH of 2.9, and the total acidity, sulfates, and total iron were 375, 3,560, and 460 ppm, respectively. The comparable figures for another acid drainage were: pH 1.4; total acidity, 7,800 ppm; sulfates, 41,700 ppm; and total iron, 12,270 ppm. Hoffert (3) has reviewed recently the status of acid mine drainage as one of the important industrial wastes.

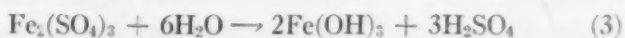
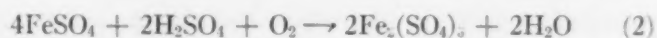
*Attempts at solving the acid mine drainage problem.* In the past, extensive programs for air sealing acid-producing abandoned coal mines have been undertaken with the purpose of reducing both the volume of the acid waters issuing from the mines and the amount of soluble acidic materials dissolved in them. It has been assumed that if the volume of surface waters gaining admission to such mines could be reduced, less of the objectionable acid drainage would be formed. It has also been assumed that if atmospheric oxygen could be cut off from the mine, or if the concentration of the oxygen present in the mine could be markedly reduced, less oxidation of the sulfur and sulfur-bearing compounds of the coal beds would result, and thus smaller amounts of acid compounds would be available for solution in the water. The following reaction indicates what is assumed to take place when the iron disulfides are oxidized:



In addition to attempts to limit the acid mine drainage at its source, work has been done on modifying the acid drainage issuing from bituminous coal mines. Chemical treatments have been studied to rid these waters of undesirable constituents or to modify their composition so that they would no longer be considered objectionable.

However, because of the cost of such chemical treatments and the unsolved problem of disposing of the large quantity of sludge which would be produced, no feasible process has been devised.

*Normal changes in acid mine drainage after its formation.* It has been assumed generally that the change of clear acid mine water to the characteristic amber or rusty color, which the water soon acquires after exposure to the air and which aids in making this type of pollution so objectionable, is due to the atmospheric oxidation of ferrous to ferric sulfate. The subsequent hydrolysis of the latter compound yields ferric hydroxide. This compound and hydrated iron oxides cause the marked rusty-colored water and precipitates in the water courses. The formation of the ferric sulfate and its hydrolysis may be shown by the following equations:



When bottles of clear acid water, taken from abandoned areas of bituminous coal mines (Pittsburgh seam) in Monongalia County, West Virginia, were stoppered so that no air space existed, the water remained clear even after weeks of storage. However, if a bottle was only half filled with water so that air was readily available, the amber color was formed within 24 to 48 hours, the deeper color being formed upon longer standing. Also, if clear acid mine water was placed in a container and carbon dioxide allowed to bubble through it, the amber color was not formed after a week; yet, if another container of the same water was exposed freely to the air, the color formation was rapid. If the passage of the carbon dioxide was stopped and that container then exposed to the air, the amber color developed in that water. In other words, air influences the characteristic change from the clear to the amber-colored water.

While it has been known that some members of the *Chlamydothales* have an iron compound in their sheath after growing in iron-bearing water, the role that microorganisms might have in inducing changes in the iron compounds of acid mine drainage appears to have been overlooked. The purpose of this preliminary report is to show that microorganisms do have an active role in the above interrelationship and to propose that the formation of sulfuric acid within the mine may be due to microbial action.

This paper is published with the approval of the directors, Agricultural Experiment Station (Scientific Paper No. 373), and Engineering Experiment Station, West Virginia University.

*The role of microorganisms in the oxidation of iron in acid mine drainage.* That the presence of air is not the sole criterion of whether the water's ferrous sulfate will change to ferric sulfate will be seen from the following simple experiments. If mercuric chloride, phenol, or formaldehyde—known disinfectants—are added to clear acid mine water in concentrations varying from one part per thousand to one part per hundred billion, the amber color starts to form in the water containing the smallest concentration of the chemical and in the control. It has been noted that for any chemical the appearance of this color varied with the length of incubation. Some of the waters containing the smaller concentrations of the chemicals may be clear after a week, but upon continued incubation they become amber. However, it was found that a concentration of one part per million of mercuric chloride, one part per thousand of phenol, and one part per ten thousand of formaldehyde prevented any formation of the amber color, irrespective of the length of holding.

In another test toluene was added to a half-filled bottle of clear acid mine water and the container closed. There was no amber color formation. Again, if clear mine water is filtered through an "E.K." germicide pad of a Seitz filter and this sterile water, in sterile cotton-stoppered containers, is exposed to the air, no amber color is produced. Too, if a synthetic mine water, made from distilled water and the iron disulfide (pyrite and marcasite) contained in the "sulfur balls" from acid-producing bituminous coal mines, is treated similarly, no amber color is produced in it after weeks of air exposure.

Thus, since the addition of bactericides such as mercuric chloride, formaldehyde, and phenol and a bacteriostatic agent such as toluene prevents the change from clear water to the amber color of these air-exposed acid mine waters, and since a Seitz-filtered water, where particles of microscopic size are removed, behave similarly, there is strong evidence that atmospheric oxygen is not the only agent involved in the formation of the undesirable hydrated oxides of iron. That this view is valid may be demonstrated by inoculating a Seitz-filtered portion of acid mine water or a Seitz-filtered portion of synthetic mine water with raw, untreated acid mine water. The characteristic amber color is then produced in the inoculated water.

To determine the nature of the microorganism or microorganisms concerned, 1 ml. of the raw water which had just turned amber was inoculated into 10 ml. of clear acid water sterilized by Seitz filtration. When this became amber, it served as the inoculum for a second tube of this sterile water. After such serial transfers had served to enrich the agent or agents present in the colored water, a portion of the liquid from the last amber tube was streaked on the surface of a poured agar plate composed of equal volumes of sterile mine water and 3 per cent agar-agar. After 6–10 days of incubation at room

temperature (22–28°C.), an amber color appeared in the agar, and very small colonies were apparent in and around the colored areas. Pure cultures of the bacteria were secured by repeated replating upon the acid-water agar. Microscopic examination of the organisms of the colonies proved the presence of gram-negative, non-sporeforming rods approximately  $0.4\ \mu$  wide by  $0.8$ – $1.0\ \mu$  long. When either sterile, raw acid mine water or synthetic acid mine water was inoculated with this bacterium, the waters became amber and formed the typical rusty precipitate, whereas uninoculated sterile control tubes of the same waters remained clear. Thus, a bacterium has been isolated in pure culture which is capable of growing in acid mine water and in bringing about the oxidation shown in Equation 2. One of the acid waters commonly used as the substrate in the tests had a pH of 2.2 and a total acidity of 7,938 ppm as calculated from its titration with N/50 NaOH in hot solution to the phenolphthalein endpoint. Attempts to grow these iron-oxidizing bacteria in the synthetic sulfur medium of Waksman and Starkey (12) (which serves as an excellent substrate for *Thiobacillus thiooxidans*, a well-known sulfur oxidizer acting at low pH) have failed. Further morphological, cultural, and physiological studies are being made on the organism.

Besides the bacterium mentioned above, several fungi have been isolated from acid mine drainage. None of these seems to be an agent causing rapid oxidation of ferrous iron to ferric iron. One of the isolates has been tentatively placed in the genus *Spicaria*.<sup>1</sup> Microscopic examination of wet mounts of it growing in this water shows a compound of iron to be precipitated on the fungal spores and mycelium. On the solid substrate of this water the characteristic amber color is formed.

*Are microorganisms involved in the formation of acid mine drainage?* Equation 2 shows the change which takes place in the oxidation state of the iron of the acid mine water after this water has been produced. The findings that microbial activity is involved in this oxidation, and that atmospheric oxidation is not the sole factor, is an interesting one, yet in itself it is a decidedly minor part in the major problem concerned in this type of industrial waste.

The major problem deals with the actual production of acid water in the mine. Several questions now arise: Is Equation 1, the accepted reaction for the formation of the offensive materials of the acid drainage, solely an atmospheric oxidation phenomenon, or are microorganisms involved there also? Are the abandoned areas of the acid-producing bituminous coal mines capable of supporting microbial growth? Are there any microorganisms known which can use the substances present in these areas in their life activities and produce, as by-products, the offending materials found in acid mine drainage?

<sup>1</sup> We are indebted to H. L. Barnett for aid in identifying this fungus.



Carpenter and Herndon (1), in studying the sterilizing effect of acid mine waters on domestic sewage, noted the growth of an organism on their plates of nutrient agar at or near a pH of 3.0 which they identified as "*Thiobacillus thiooxidans* or a very closely related species." None of the tests used by them to arrive at such a conclusion was given. Their attempts to find the organism responsible for the acidity of the acid mine water were unsuccessful. Various coals were examined by these workers for the possible role of microorganisms in the change of the sulfur content. By comparing sterilized with nonsterilized portions they concluded that at least part of the sulfate produced in acid mine water can be attributed to bacteriological action.

In our laboratory some exploratory tests made on acid drainage have given interesting results with regard to the possible significance of bacteria in the formation of the sulfuric acid in the mine. Although numerous water samples have been studied, the sources of four typical samples are given. In one instance the water sample was taken outside of the mine from the flume into which pumps had emptied the drainage taken from an abandoned area several hundred feet below the earth's surface. In another the water was taken inside the mine as it ran out of an abandoned area and flowed along the main haulage way. A third sample came directly from an abandoned area and was caught as it issued from a bore hole in the coal. A fourth was taken from a strip mine where the water was running out of a spoil bank.

All the waters were inoculated into the synthetic medium of Waksman and Starkey (12), used for the culturing of *Th. thiooxidans*.<sup>2</sup> Growth appeared in all the inoculated flasks within 7 to 10 days, and, at that time, subcultures were made into fresh media. Repeated transfers have always yielded an active culture. Material from the flasks was streaked on the surface of the solid sodium thiosulfate medium of Waksman (11), and minute straw-yellow to cream-colored colonies were produced. Thus, a second bacterium has been isolated from acid mine drainage. Whereas the first bacterium was instrumental in the change of ferrous to ferric iron, attempts to grow it in the sulfur medium have failed. The second, however, will rapidly oxidize sulfur to form sulfuric acid, but attempts to have it oxidize the ferrous iron of the acid water have failed.

Growth taken from the liquid or solid substrate was morphologically similar to the published descriptions of *Th. thiooxidans*. Physiologically, the organism fits the description of the known sulfur oxidizer. It can satisfy its carbon requirements from the carbon dioxide of the air; its nitrogen can come from ammonium sulfate; and its energy can be derived from the oxidation of elemental sulfur or thiosulfate. The pH of the liquid medium varies from 1.5 to 0.9 after one to two weeks of growth.

<sup>2</sup> A pure culture of *Th. thiooxidans* was obtained from R. L. Starkey, to whom we are indebted for this favor.

*Consideration of environment where acid mine drainage is produced with reported activities of Th. thiooxidans.* It is felt that since all of the typical acid mine waters sampled have yielded organisms which fit the description of *Th. thiooxidans*, it is worth while to consider the possibility of its acting as the agent in the formation of sulfuric acid in the abandoned areas of bituminous coal mines. If one considers the actual environmental conditions existing in such areas, the possibility of this organism functioning there is quite striking.

Two gas analyses, made on abandoned areas from which one of the samples of water was taken, showed the absence of carbon monoxide in both. The methane values were 12.4 and 18 per cent; the oxygen concentrations were 3.2 and 9.4 per cent; whereas the carbon dioxide values were 3.5 and 3.8 per cent. The work of Vogler and Umbreit (10) indicates that *Th. thiooxidans* would not find some of these gaseous components of this area prohibitive to its action. These workers ran experiments using pure cultures of *Th. thiooxidans* in which the gaseous environment varied from 10 to 30 per cent of oxygen and from 0.01 to 10 per cent of carbon dioxide. By using the sulfate sulfur, formed by the oxidation of elemental sulfur, as a measure of the activity of the organisms under the experimental conditions, they felt their data showed that "a small amount of variation is evident, but that variation bears no consistent relation to either oxygen or carbon dioxide pressures. Significantly, the oxygen can be decreased to half its value in air without detectable influence upon the rate of sulfate formation." A study of the work of Vogler (8) brings out the striking ability of this organism to modify its activities under the influence of carbon dioxide. Indeed, he postulates that the "carbon dioxide may act as an oxidizing agent for sulfur."

It is well known that the waters issuing from these abandoned areas of acid producing mines are quite acid and have a high sulfate content. Waksman and Starkey (12) long ago called attention to the unusual property possessed by *Th. thiooxidans* of being able to grow when the medium contained 0.5 M concentration of sulfuric acid, equivalent to nearly 5 per cent concentration of the acid. Starkey (6) found that oxidation proceeded most rapidly between pH 2.5 and 3.5, although oxidation of sulfur is very rapid over a very much wider range. Above 6.0 there is practically no oxidation of sulfur in the culture medium by *Th. thiooxidans*. Vogler, LePage, and Umbreit (9) reported that above a pH of 5.0 there is a marked inhibition of sulfur (and thiosulfate) oxidation, but there is little effect upon the endogenous respiration in the absence of sulfur. These workers felt that the organism is adapted to just that range of H-ion concentration in which free carbon dioxide is dissolved in the medium as a gas. Waksman and Starkey (12) and Starkey (6) have noted that *Th. thiooxidans* is quite tolerant to the sulfate ion. The latter states: "However, the end product of sulfur oxidation (sulfate) is tolerated by

*Thiobacillus thiooxidans* in as high concentration as 20 per cent in the form of magnesium sulfate."

The temperature of the acid waters issuing from the abandoned areas tends to be below 20°C. Vogler, LePage, and Umbreit (9) found that sulfur oxidation proceeded over a temperature range of 7° to 45°C.

Thiessen (7) reported that sulfur in all coals is "present in amounts ranging from traces to as high as 10 or more percent, commercial coals of the Eastern United States contains from 0.5 to 1.5 percent of sulfur." Thiessen stated that sulfur does not occur as such in coal but is present in organic combination as part of the coal substance and in inorganic combination as pyrite or marcasite and, especially in weathered coals, as calcium sulfate. Mellor's (5) compilation is a rich source of information dealing with the iron disulfides, pyrite and marcasite. Two of the many items of interest concerning these inorganic combinations of iron and sulfur are the statements concerning the weathering or the ready breakdown of marcasite under the influence of moist conditions and the finding of free sulfur in the naturally-occurring iron disulfides.

Kirner (4) reports that bituminous coal has a nitrogen composition ranging from 0.6 to 2.8 per cent. The complete story of the forms in which this nitrogen may appear in the coals and, indeed, whether such compounds are available to microorganisms is not known. Starkey (6) found that with *Th. thiooxidans* there is some oxidation in media receiving no nitrogen other than that which may have been absorbed from the air or introduced as impurity in the non-nitrogenous compounds incorporated in the medium. Of the nitrogen sources investi-

gated, ammonium nitrogen is the only one that has been found to be available to the organism.

In this work, a bacterium, unidentified as yet, has been found in acid mine drainage which is involved in the oxidation of ferrous to ferric sulfate. A second bacterium similar, if not identical, in its morphological, cultural, and physiological characters to *Th. thiooxidans* has been isolated repeatedly from the acid mine drainage of some bituminous coal mines. It is postulated that this latter or an unknown similar organism is involved in the oxidation of the sulfur and the sulfur compounds to sulfuric acid. The increasing demands for abatement of stream pollution by acid mine drainage has intensified the efforts of many agencies to solve the problem. It is felt that the mine-sealing program has not given a complete solution and it is suggested that the study of the relation of certain microorganisms to the formation of the sulfuric acid in the mine might be a most profitable one.

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## Reconnaissance Soil Survey Work in Japan

C. L. W. Swanson

Department of Soils, Connecticut Agricultural Experiment Station,  
New Haven, Connecticut

THE RECONNAISSANCE SOIL SURVEY OF Japan now being conducted by American scientists is the first of its kind to be made of that country using modern survey methods for classifying and mapping soils in terms of their morphological features. Soil maps of Japan now available from Japanese sources have been made according to the old geological method of soil surveying similar to that used initially in the United States.

During the past 20 years Japan has produced only 80-85 per cent of her annual food requirements. Because of the food shortage there and in the world as a whole, the maintenance or increase of food production has now

assumed particular importance. Since modern soil maps which could be used as an aid in maximizing Japan's food production were unavailable, the survey was undertaken.

The most important agricultural areas are being mapped first, particularly the larger plains and more nearly level regions. The hilly and mountainous sections are of little importance agriculturally. A preliminary field survey was made of Japan before any reconnaissance surveys were initiated. Reconnaissance soil surveys of the Kanto Plain area near Tokyo and the island of Kyushu have been completed (Table 1). Surveys in progress include the islands of Hokkaido and Shikoku, and the Osaka Plain on the main island of Honshu. A soil map (1:250,000) printed in color and a report similar in outline to that of the Soil Survey Division, Bureau of Plant Industry, Soils, and Agricultural Engineering, U. S. De-

<sup>1</sup> Formerly Major, Air Corps, and head of the Soils and Fertilizer Branch, Agriculture Division, Natural Resources Section, General Headquarters, Supreme Commander for the Allied Powers, Tokyo, Japan.



Department of Agriculture, is to be published for each area surveyed. Because of the nature of the survey, only associations of soils are shown on the soil maps.

Several sources of data and information are used in preparing the map and report. Much of the information obtained through field observation of the soils by experienced American soil surveyors and from Japanese soil and geological maps. Map information and soil data already gathered by Japanese scientists are being coordinated and interpreted in terms of modern soil concepts and nomenclature. Arrangements have been made with the Soils Departments of the Imperial Universities of Japan to make physical and chemical determinations on soil samples taken in the field during the course of the survey. Samples are also being sent to the Soil Survey Division, U. S. Department of Agriculture, for analysis.

TABLE 1

Soil group	Kanto Plain*		Kyushu†	
	Acres	Percent of total area	Acres	Percent of total area
Brown forest.....	2,241,551	28.14	777,431	8.51
Alluvial.....	1,605,317	20.16	981,531	10.75
Black forest.....	100,332	1.26	115,275	1.26
Half bog.....	75,647	0.95	0	0
Planosols.....	35,036	0.44	224,599	2.46
Red and yellow podzolic.....	0	0	193,689	2.12
Lithosols.....	691,975	8.69	1,829,257	20.04
Woods.....	73,258	0.92	34,621	0.38
Rough mountainous land.....	2,981,302	37.44	4,941,461	54.11
Barren volcanic rock.....	0	0	33,909	0.37
Water.....	159,258	2.00	0	0
Total.....	7,963,676	100.00	9,131,773	100.00

\*About 33 per cent of the area is now being used for agricultural purposes.  
†About 20 per cent of the area is now being used for agricultural purposes.

tation in soil development, establishing the morphology of the soils themselves as the basis of classification. Detailed soil surveys of parts of Aomori and Gifu Prefectures were made by the Japanese using this classification scheme, the soil survey program having been curtailed during World War II. The Japanese are now being encouraged to activate their soil survey work as rapidly as possible. Whenever practicable, technical assistance in the reactivation of their soil survey program is being given by American soil scientists. In this connection, the published reconnaissance soil survey reports and maps of Japan will be made available to them.

Certain representatives of the major zonal and intrazonal soil groups of the world are now being identified in Japan (Table 1 and Fig. 1). It is being observed that profile characteristics are similar to those in the United



FIG. 1. Generalized soil map of Japan.

States at comparable latitudes; it appears, however, that local topography and parent material, especially volcanic ash materials, exert more influence on the kinds of soils developed than does climate. When one considers the extremely rugged terrain and steep slopes that prevail over most of Japan, it is understandable why the Japanese, until recently, gave little consideration to climate and vegetation influences as important soil-forming factors. The many diastrophic movements that have occurred quite recently, geologically speaking, have naturally tended to maintain or increase the areas of rugged terrain and steep slopes, thereby continuing conditions conducive to the development of immature and young soils. The high rainfall generally prevalent in Japan helps to hinder the development of soil profiles by eroding weathered materials from the slopes. The constant changing of the topography, the continual removal of weathered materials to lower levels, the additions of volcanic ash, and the high rainfall have all combined to assure

For many years Japanese soil scientists believed that the soils of the country did not fit into the prevailing scientific scheme of classification and, therefore, would not accept the modern concept of great climatic soil groups. While placing their greatest emphasis on geological effects on the formation of soils, they did admit that vegetation and climate exerted certain influences on soil formation processes. It is not surprising, however, that the Japanese did not readily accept modern soil survey methods, for even in the United States there existed certain areas where initially it was difficult to apply the modern pattern. As late as 1935, Marbut, in his *Soils of the United States* (see C. B. Hutchison's *California agriculture*. Berkeley: Univ. California Press, 1946), was unable to delineate in California representatives of the great soil groups of the world.

In 1937, however, an Outline Soil Map of Japan (1:5,000,000), prepared by Toyotaro Seki, of the Imperial Agricultural Experiment Station in Nishigahara, Tokyo Prefecture, was published in an article by L. G. Scheidl (*Mitteil. dtsch. Ges. Natur Volk. Ost.*, Vol. 30, Pt. A). This map includes and recognizes the role of climate and vege-

that materials in a given locality would not be given time to develop mature profile characteristics.

The most important soils of Japan, which support the bulk of the population, are the alluvial soils developed in the lowlands. On these, paddy or irrigated rice is grown during the summer, and in southern Japan (Fig. 2) during



FIG. 2. Northern limits of a number of crops in Japan.

the winter months these soils are also cropped to wheat or barley. Many of the important agricultural soils are developed on volcanic ash (Fig. 1). Brown forest and black forest soils,<sup>2</sup> derived principally from volcanic ash materials, are important upland soils used for crop production purposes. Since these are especially low in available phosphorus, they are comparatively low in inherent fertility. They are physically well suited to growing small grains, white and sweet potatoes, vegetables, mulberry, upland rice, tree fruits, tea, tobacco, and other non-irrigated crops.

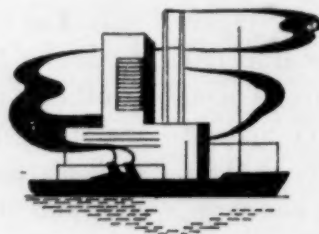
In many of the red and yellow podzolic soil areas (Fig. 1), because the demand for cropland is so great, a large

<sup>2</sup> The names "brown forest" and "black forest" were originally given to the soil groups so designated by Japanese and German soil scientists and some question exists as to the correctness of this application. The brown forest soils do not have all of the profile characteristics normally associated with brown forest soils, being acidic in nature and slightly podzolized. Consideration is now being given to assigning a new great soil group name to these soils.

number of the steep slopes poorly suited to agriculture are terraced and cropped. The crops grown are wheat, barley, upland rice, white and sweet potatoes, vegetables and, in many places on southern slopes facing the sea, citrus fruits are grown. Podzol soils occur at the higher elevations in Hokkaido and northern Honshu, but they are more widespread in the former area than in the latter. In these areas they are associated with coniferous vegetation. Most of the gray-brown podzolic soils, which are developed in the hill and mountain areas of the podzols (Fig. 1), are forested, but where the soil is deep enough, farmers have either terraced them or planted the row crops up and down hill without the benefit of terraces. In the untterraced fields, soil erosion is quite severe. White potatoes, tree fruits, vegetables, small grains (wheat, barley, oats), corn, and hay are the upland crops grown on these soils. Lithosols, which are found on the perimeter of the undulating and rolling plains adjacent to mountains and in upland areas in mountainous regions, are not well suited to crop production and are mostly in forest vegetation.

For maximum crop yields, most of the soils of Japan require heavy fertilization and liming. Fertilizers are of real importance because of the intensive agriculture and because the scarcity of agricultural land makes it important to secure high yields from every crop grown. The preponderance of hilly and mountainous terrain sets definite limits to the cultivated area and explains why no more than 15,000,000 acres, or 16 per cent of the total area of Japan, is cultivated.

Locally produced materials, such as compost, night soil (human excrement), green manure, and wood and straw ashes are used extensively as fertilizer materials. Bean cake and fish fertilizers have been applied to the soil in the past, but recently these materials have been used largely for food purposes. Large quantities of commercial fertilizers, particularly ammonium sulfate and superphosphate, have been employed. Their use was curtailed during World War II because production of nitrogenous fertilizer was retarded, while production of phosphatic fertilizer was dependent on imports of phosphate rock. When world consumption (1935-37) of commercial fertilizers used by each country is considered, Japan ranked third for nitrogenous fertilizers and sixth for phosphatic and potassic fertilizers (K. G. Clark and M. S. Sherman, *Prewar world production and consumption of plant foods in fertilizers*. U. S. Dept. Agric. Misc. Publ. 593, 1946). Yet that country ranked nineteenth in the world distribution of arable lands.





# NEWS and Notes

## Large-scale extraction of fossils

from limestone rock by an etching process now in progress at the Smithsonian Institution is expected to yield over 1,000,000 new specimens for invertebrate collections. The rock bearing the fossils is immersed in muriatic acid in large, acid-resistant vats, the limestone being thus dissolved to yield the silicified fossils intact. Over the last 8 years more than 12 tons of limestone have been brought to Washington, D. C., from the rich Permian deposits of West Texas. It is estimated that it will require 3 years to secure the fossils from the 8 tons of rock remaining to be processed, but at the end of this time the Smithsonian paleontologists expect to find 700-1,000 species, many of which will be new.

According to G. A. Cooper, who is directing the classification of brachiopods, an estimated 50 per cent of all the fossil brachiopods recovered will be new specimens. This class, which is distinguished by its inequivalve and bilaterally symmetrical character, appeared first during the Devonian period, reaching its highest differentiation and greatest distribution during the Permian. At the present time there are only about 200 known species and 60 known genera, distributed widely over marine areas, but abundantly in only a few.

## About People

**Maximilian Philip**, chairman, Department of Mathematics, City College of New York, retired September 1 after 45 years in that Department. Prof. Philip is succeeded by **Warren G. Hubert**, who has served on the City College staff since 1912.

**Jesse W. Huckert**, formerly associate editor of *Product Engineering*, has

been appointed to the staff of the Department of Mechanical Engineering, Ohio State University, and will assume his duties there October 1.

**E. L. Porter**, professor of physiology, University of Texas Medical Branch, Galveston, is spending several months with **H. S. Liddell**, professor of psychology, Cornell University, and director, Cornell Behavior Farm, where both will continue their joint studies on chronic experimental neuroses in animals.

**D. P. Costello**, professor of zoology, University of North Carolina, has been appointed chairman, Department of Zoology. **R. E. Coker**, former chairman, has resigned to continue as Kenan professor of zoology for teaching and research.

**Carleton Shugg**, general manager of Todd Shipyards Corporation in charge of New York Harbor work, has been appointed manager, Hanford Directed Operations, U. S. Atomic Energy Commission, with offices at Richland, Washington.

Among those retiring from the staff of Ohio State University this summer with the rank of professor emeritus were: **John F. Cunningham**, dean, College of Agriculture; **Clyde T. Morris**, chairman, Department of Civil Engineering; **Leonard W. Goss**, chairman, Department of Veterinary Pathology; and **Carl W. Gay**, professor, Department of Animal Husbandry.

**Vernon E. Krah**, formerly of the Department of Anatomy, Wayne University College of Medicine, has been appointed associate professor, Department of Gross Anatomy, University of Maryland School of Medicine, Baltimore.

**R. Blackwell Smith, Jr.**, formerly of the Pharmacological Department, Food and Drug Administration, Washington, D. C., on July 1 assumed the deanship of the School of Pharmacy, Medical College of Virginia, Richmond.

**David W. Bishop**, formerly assistant professor of zoology, University of Colorado, and visiting lecturer, University of Illinois, during the current summer session, has been appointed to the staff of the Department of Zoology, University of Illinois.

**Robert H. Cotton**, director, Plymouth Division, National Research Corporation, has resigned to become supervisory chemist at the Citrus Experiment Station, University of Florida, Lake Alfred.

**Curt Stern** has been appointed professor of zoology at the University of California, Berkeley, and **William E. Berg** and **Oliver P. Pearson** have been made instructors in the Department.

**Chester A. Darling**, retired professor of biology, has been appointed acting president of Allegheny College, Meadville, Pennsylvania, and will serve until a permanent head is selected to succeed **John R. Schultz**, 14th president of the College, who died August 11.

**Solon W. Stone**, teaching fellow at Harvard University, has been appointed instructor in geology at Trinity College, Hartford, Connecticut.

**Harold D. Webb**, one of the scientists who established radar contact with the moon in January 1946, has been appointed assistant professor of electrical engineering in the University of Illinois College of Engineering. Since 1942 Dr. Webb has been employed at the Army Signal Corps, Belmar, New Jersey.

**Walter L. Stutz**, chief of the Engineering Instruments and Mechanical Appliances Section since 1928, has retired from the National Bureau of Standards after 35 years service. As chief of the Section, Dr. Stutz has been principally concerned with the direction of testing activities for other government agencies, including devices relating to flood control, power development, irrigation, and soil conservation.

**Arthur Dorne** and **Joseph Margolin**, formerly of the Antenna Design Section of the Airborne Instruments Laboratory, Mineola, New York, have established an antenna consulting firm—Dorne and Margolin, Antenna Consultants—effective on September 1, with offices temporarily located at 126 North Ocean Avenue, Freeport, Long Island, New York. While at the Airborne Instruments Laboratory, the two men were largely concerned with the faired-in, dragless antenna designing groups.

**T. G. Yuncker**, head, Botany Department, DePauw University, and during 1947 a visiting research professor at the University of Illinois, returned to DePauw late in August, having brought his studies of the Piperaceae to a close.

**Grant W. Smith** has resigned as research chemist and supervisor, Department of Polymerization Research, B. F. Goodrich Company, Akron, Ohio, to be-

come professor and head, Department of Chemistry, University of North Dakota, Grand Forks.

**John P. Buwalda**, professor of geology, retired as chairman of the Division of Geology, California Institute of Technology, on July 1, in order to complete a number of research projects in structural geology upon which he has been working for some time. He will continue as professor in the Division. **Chester Stock**, professor of paleontology, succeeded Dr. Buwalda as head of the Division.

### Visitors to U. S.

**H. F. Frommurze**, principal geologist of the Geological Survey of the Union of South Africa, is in the United States to study methods used by the U. S. Geological Survey in the exploration of ground water.

**Yngve Andersson**, research agricultural engineer at the Swedish Institute of Agricultural Engineering, Uppsala, recently visited Iowa State College, Ames, on a five-month tour of the United States. Dr. Andersson will investigate methods used in experiment stations, extension service methods, and use of machinery on farms.

**Francis V. Groër**, former professor of pediatrics and head, Pediatric Department, University of Lvov, now in Cracow (Poland), has been visiting the United States as Polish delegate to the Nutritional Committee of the Children's Save Fund of the UNO and to the International Congress of Pediatrics, held recently in New York City. Dr. Groër was Theodore B. Sachs visiting professor at the University of Illinois, Chicago, and Hanna lecturer at Western Reserve University, Cleveland, in 1931.

### Fellowships

The Institute of International Education has been requested by the Argentine National Cultural Commission to assist in selecting two U. S. graduate students who will receive fellowships from the Commission for 10 months study in Argentina. Laurence Duggan, director of the Institute, has announced that these fellowships, running from March 1 to December 31, 1948, will provide round-trip travel plus about \$1,250. Applicants must be research workers of recognized standing in the arts, humanities, natural

sciences, or social sciences; their fellowship work must be of positive benefit to the cultural development of the U. S. and Argentina; they must have a working knowledge of Spanish; and they must be between 25 and 45. Details and forms may be obtained from the Institute at 2 West 45th Street, New York 27. The closing date is October 31.

The **Julius Rosenwald Fund** is offering fellowships to average about \$2,000 each to Negroes in any part of the United States, and to white Southerners who wish to work on some problem distinctive to the South and who expect to make their careers in the South. The fellowships are open to men and women between the ages of 24 and 35 who have completed their general college or professional course before making application. The term of the fellowship will be for one year, and grants will not be awarded for a term of less than six months. Applicants are not restricted to any special subject or activity. This is the last year that Rosenwald Fellowships will be available, since the Fund is closing its work next June. The closing date for applications is January 1, 1948. Application blanks may be secured from Mrs. Hilde Reitzes, Committee on Fellowships, Julius Rosenwald Fund, 4901 Ellis Avenue, Chicago 15, Illinois.

### Grants and Awards

The **Gold Medal of the American Society for Metals** will be awarded in October to **C. H. Mathewson**, professor of metallurgy, Yale University. Dr. Mathewson, who is known for his work on constitution of alloys, crystallography, and recrystallization of metals, receives the medal "for outstanding metallurgical knowledge and great versatility in the application of science to the metal industry, as well as exceptional ability in the diagnosis and solution of diversified metallurgical problems."

The **Inter-American Institute of Agricultural Sciences** at Turrialba, Costa Rica, has recently received several grants. The Standard Oil Development Company has transferred the sum of \$9,500 as the first of a series of annual payments to meet the cost of fellowships to provide for researches and investigations on the more economical and more efficient production of tropical crops of major importance, with particular refer-

ence to the application of chemical petroleum by-products for the destruction of weeds and also for the control of growth of certain plants of economic value. Standard Oil will also meet the expense of a visiting agricultural scientist, **Orin Smith**, Cornell University, who will spend three months a year at the Institute, outlining and directing the work of the Fellows. The American Coca Research Committee, Washington, D. C., has transferred \$50,000 to the Institute as a contribution to the cost of conducting an intensive research that will reduce the existing high percentage of crop failures and develop new techniques in the planting and cultivation of cacao. The American International Association for Economic and Social Development has granted the Institute \$158,000 to permit an extension of research and educational facilities there, particularly in the Animal Industry Department. The grant will also permit Institute scientists to train students and farmers selected to become teachers and demonstrators on the staff of the Association.

The **Inter-American Institute of Agricultural Sciences**, whose executive offices are in the Pan American Union Building, Washington, D. C., operates on the basis of a Convention by which the governments of the American republics have agreed to recognize the permanent status of the Institute as an inter-American entity. Its purpose is to encourage and advance the development of agricultural sciences in the American republics through research, teaching, and extension activities in the theory and practice of agriculture and related fields. The Institute is divided into four major departments, Plant Industry, Animal Industry, Agricultural Engineering, and Agricultural Economics and Rural Welfare, and is at present engaged in research work on important tropical industrial and food plants as well as animal industry.

The Institute is governed by a Board of Directors, made up of one representative from each American Republic, whose chairman is **Antonio Rocha**, of Colombia. Members of the Administrative Committee include: **H. Harold Hume**, provost, College of Agriculture, University of Florida; **Robert Earle Buchanan**, director, Agricultural Experiment Station, Iowa State College of Agriculture and Mechanical Arts; and **Luis Cruz**, San José, Costa Rica. Director of the Institute is **Ralph H. Allee**, Costa Rica, and secretary is **José L. Colom**, Washington, D. C.



Research looking toward the decrease in deaths from gangrene diseases will be continued by the Department of Bacteriology, Indiana University, as the result of an annual grant of \$13,860 from the National Institute of Health. The research, initiated last year, is being conducted by L. S. McClung, head of the Department, in cooperation with Harry G. Day, chemist.

## Colleges and Universities

The Department of Anthropology, University of Colorado, has appointed Robert H. Lister, Harvard University, to its staff. Robert F. Burgh, an archaeologist, has been appointed assistant to the director.

The Biology Department, University of Notre Dame, has recently made several staff changes and appointments. Donald J. Plunkett, associate professor of biology, has been named assistant head of the Department; John A. Jump has been promoted to associate professor of botany; Noe Higinbotham, assistant professor of botany, has been granted leave of absence to participate in radio-biology research at the Argonne National Laboratory, Chicago; Albert L. Delisle, formerly director, Botanical Division, Universidad Nacional Facultad de Agronomía, Medellin, Colombia, has been appointed associate professor of botany and curator of the University's Herbarium; Louis E. DeLanney, San Jose Teachers College, has been named assistant professor of embryology; and Edward O. Dodson, formerly of Dominican College of San Rafael, will become instructor of vertebrate zoology.

The Department of Zoology and Entomology, University of Tennessee, has added three new assistant professors to its staff during the past academic year: John M. Carpenter, of the University of Texas; Joseph C. Howell, Cornell University; and James T. Tanner, Cornell University.

The Department of Physics, Illinois Institute of Technology, has announced the promotion of W. E. Bennett from associate professor to professor, R. G. Nuckolls from assistant to associate professor, and A. G. Meister from instructor to assistant professor, and the appointment of F. F. Cleveland and F. L. Yost as professor and associate professor, respectively.

The University of Miami, Florida, has announced the following additions and promotions involving members of the Department of Zoology: Jay F. W. Pearson, to vice-president of the University; E. Morton Miller, to chairman of the Department; Julian C. Corrington and Henry F. Strohecker, to professors. New associate professors are Lauren C. Gilman and Luis René Rivas, and Vernon Gregory has been named instructor. F. G. W. Smith is director of the Marine Laboratory.

Boston College has made the following appointments in its Department of Chemistry: Samuel Glasstone, wartime consultant in electrochemistry, Zenith Radio Corporation, and formerly professor of chemistry, University of Oklahoma, professor; Andre de Béthune, Massachusetts Institute of Technology, and Timothy McCarthy, Massachusetts State College, Devens, assistant professors; and Albert F. McGuinn, S. J., re-appointed head of the Department upon the resignation of John K. Rouleau, who has held this position for the past year.

At Kent State University the Physical Science Department has been divided into the Departments of Chemistry and Physics. Will Thompson, formerly head of the combined departments, is now head of the Department of Chemistry, and G. K. Schoepfle, of the Department of Physics.

The University of Cincinnati Research Foundation, under a \$67,000 contract with the U. S. Army Air Forces, has begun a study of atmospheric conditions to discover the nature and properties of haze. Boris Podolsky, associate professor of mathematical physics, will direct the work, assisted by Roy J. Kennedy, experimental physicist, U. S. Bureau of Standards.

The Ohio State University Research Foundation is at present engaged in a two-year research project under contract with the Army Signal Corps to investigate the effects of arctic temperatures on electric batteries. Data collected from experiments, directed toward finding the most practicable solvent for use in batteries at temperatures reaching  $-100^{\circ}$  F., have shown water solutions of alcohol, hydrochloric acid, fluoroboric acid, and perchloric acid to be most satisfactory. Studies on electrolytes were then made

to discover the best conducting medium for reducing the high internal resistance of the battery under extremely low temperatures. The investigators are now attempting to determine the most suitable electrochemical couple, or proper combination of chemicals necessary to produce electricity.

A planned program of dental research which will include studies on the bacteriological and biochemical aspects of dental caries, with special emphasis on the possible importance of proteolytic bacteria in the causation of caries, is to be undertaken at the University of Rochester School of Medicine and Dentistry under a contract with the Army Medical Research and Development Board. The investigation, to be conducted in the Departments of Bacteriology and Dental Research under the direction of George P. Berry and J. E. Gilda, will be carried on in close cooperation with Basil G. Bibby, director of the Eastman Dental Dispensary. One or more selected Dental Corps officers will be assigned to participate in this and subsequent studies and to do graduate work in the biological sciences. Capt. George W. Burnett, DC, is the first officer to be so assigned.

## Industrial Laboratories

Eastman Kodak Company has recently named Cyril J. Staud new director of the Kodak Research Laboratory and John A. Leermakers assistant director and head of the photographic theory department. C. E. K. Mees, director of the laboratories since 1912, will continue to serve as vice-president in charge of research. Dr. Staud, an organic chemist, has been serving as acting director since 1943 in the absence of Dr. Mees, and Dr. Leermakers was formerly technical assistant to the director. Other appointments include: Samuel W. Davidson, business manager; Walter Clark, head of the black-and-white photography department; W. O. Kenyon, head of the high polymer department; and H. C. Yutzy, head of the emulsion research department.

M. K. Ryan, Jr., manager of the technical service section, Acetate Division, Du Pont Company, has been made manager of the fabric development section of the Division, succeeding Carl I. Taber, who was recently transferred to the Nylon Division. Mr. Ryan's position will be assumed by G. S. Hooper,

technical superintendent of the acetate yarn plant at Waynesboro, Virginia.

**Clifford S. Leonard**, formerly assistant professor of pharmacology, College of Medicine, University of Vermont, has been appointed chief, Biological Division, Lakeside Laboratories, Inc., Milwaukee, Wisconsin.

**W. Keith McAfee** has been elected chairman of the Board of Universal Sanitary Manufacturing Company, New Castle, Pennsylvania. He will also continue to give his personal attention to engineering, research, and development.

**Almon G. Hovey**, formerly in charge of the New Chemicals Development Section, is now head of the Technical Sales Service, Chemical Division, General Mills, Inc., Minneapolis.

**Albert B. Scott**, director of research and development, Norwich Pharmacal Company, and director, Eaton Laboratories, has recently resigned. Dr. Scott, who is credited with the discovery and development of Furacin, has been with the Norwich Company since 1939.

**Warren H. Goss**, assistant to the director, and head, Market and Product Development, Northern Regional Research Laboratory, U. S. Department of Agriculture, Peoria, Illinois, has been appointed associate director, Research and Products Development Department, Pillsbury Mills, Inc., Minneapolis.

**W. H. Stark**, director of research, Joseph E. Seagram & Sons, Inc., Louisville, Kentucky, has been appointed general manager, Vickers-Vulcan Process Engineering Company, Ltd., Montreal, Canada. Dr. Stark is succeeded as director of research by **Paul J. Kolachov**, previously technical counselor.

**Gale Boxill**, who was recently graduated from Washington-Lee University, has joined the Pharmacology Department, Wm. S. Merrell Company, Cincinnati, Ohio.

**Donald A. Quarles**, director of apparatus development, Bell Telephone Laboratories, has recently been elected a vice-president. He has been associated with the Bell System since 1919.

**G. F. D'Alelio**, manager, High Polymer Research Laboratory, Industrial Rayon Corporation of Cleveland, has been appointed assistant director of research, Koppers Company, Inc., Pittsburgh.

**H. B. H. Cooper**, divisional engineer associated with the Calco Chemical Division, American Cyanamid Company, Bound Brook, New Jersey, has been appointed assistant manager of the Development Department.

## Meetings

A symposium on refractory oxides has been arranged by the Refractories Division, Harbison-Walker Refractories Company, for October 10-11 at the Bedford Springs Hotel, Bedford, Pennsylvania. The program is open to all those interested in this field. The technical session, to be held on the afternoon of October 10, will include "Introduction to the Study of the Refractory Oxides," Raymond E. Birch, Harbison-Walker; "Simple Oxide Porcelains for Jet Planes and Projectiles," R. F. Geller, National Bureau of Standards; "Properties and Uses of Mullite and Pure Alumina Refractories," G. B. Remmey, Richard C. Remmey Sons Company; "Zircon and Zirconia Refractories," W. J. Baldwin, Titanium Alloy Manufacturing Company; and "Carbon as a Refractory Material," F. B. Thatcher, Great Lakes Carbon Corporation. Further information may be obtained from Collin Hyde, Harbison-Walker Refractories Company, Hays Laboratory, Pittsburgh 7, Pennsylvania.

The Highway Research Board will hold its 27th annual meeting December 2-5 at the building of the National Academy of Sciences and National Research Council, 2101 Constitution Avenue, Washington, D. C. During the week of the meeting the 6 Departments of the Board—Economics, Finance, and Administration; Design; Materials and Construction; Maintenance; Traffic and Operations; and Soils—will present and discuss important phases of highway technical development. Many of their 65 project committees will also meet.

The 6th Saranac Symposium will be held at The Saranac Laboratory, Saranac Lake, New York, September 29-October 3, inclusive. The primary topic for discussion concerns beryllium, which is assuming great importance in several lines of manufacture, with Monday devoted to its industrial phases and Tuesday, Wednesday, and Thursday morning to its medical, toxicological, and experimental aspects. Thursday afternoon is reserved for the clinical, chemical,

and engineering discussions of Shaver's disease, and the agenda for the final sessions on Friday comprise the current viewpoints on workmen's compensation for pulmonary and other occupational disease. Participants will include representatives of industrial, labor, and insurance groups, universities and research foundations, national and state health agencies and labor departments, as well as state compensation boards.

The New York Section, American Association of Cereal Chemists, opens its 1947-48 season on October 7 with a combined meeting with the New York group of the American Society of Brewing Chemists at the Building Trades Employers Association club rooms, 2 Park Avenue. Reservations for the dinner at 6:30 P. M. must be made with Lawrence Atkin, Wallerstein Laboratories, 180 Madison Avenue, New York 16. Aage Lund, chief chemist of the Tuborg breweries, Copenhagen, and co-author of the 1939 edition of Jorgensen's *Microorganisms and fermentation*, will speak on "Biological Control and Microorganisms in Breweries." Nonmembers interested in the fermentation industries are cordially invited to make reservations for the dinner or to attend the lecture, which will begin about 8 P. M.

The third annual meeting of the National Conference on Industrial Hydraulics will be held October 16-17, at the Hotel Continental, Chicago. The Conference is sponsored by Armour Research Foundation and the Graduate School, Illinois Institute of Technology, in cooperation with the Western Society of Engineers and the Chicago sections of the Society of Automotive Engineers, American Society of Civil Engineers, and the American Society of Mechanical Engineers. The following program has been planned: October 16, "Fundamentals of Hydraulic Governor Operation," G. Forrest Drake, Woodward Governor Company, Rockford, Illinois; "Hydraulic Circuits of Industrial Process Controls," H. Ziebolz, Askania Regulator Company, Chicago; "Fundamental Aspects of Cavitation," Hunter Rouse, Iowa Institute of Hydraulic Research, State University of Iowa; "Cavitation, the Limiting Factor in Marine Propulsion," Karl E. Schoenherr, College of Engineering, University of Notre Dame; "Cavitation in Centrifugal Pumps—Some of the Less Well-Known Factors," R. M. Watson, Worthington Pump and Machinery Corporation, Har-



Shaver's the final the current compensation occupational include repre- and in- d research te health s, as well

on, New Jersey; October 17, "Hydro-  
etic Transmissions," W. F. Shurts,  
in Disc Clutch Company, Rockford,  
nois; "Torque Converters: Making  
efficiency Pay Dividends," Robert  
psley, Clark Equipment Company,  
changan, Michigan; "Some Recent De-  
velopments in Hydraulic Transmissions,"  
H. Hensleigh, Consulting Engineer,  
etroit, Michigan; "Variable Speed Hy-  
draulic Power Transmission," James Rob-  
son, Vickers, Inc., Detroit, Michigan;  
Hydraulic Fluids and Their Applica-  
ons," James I. Clower, University of  
elaware; "Development of Hydraulic  
als," W. W. Hagerty, University of  
Michigan; and "Postwar Aspects and  
blems of Engineering Research," Boris  
Bakhtmeteff, Columbia University.  
urther information may be obtained  
om Le Van Griffis, Armour Research  
oundation, Technology Center, Chicago  
Illinois.

The Physical Society and the Insti-  
ute of Physics of England are jointly  
ranging a series of meetings and other  
ctions to take place September 25-26,  
1947 in London, to mark the 50th anni-  
versary of the discovery of the electron  
by Sir J. J. Thomson. A special exhibition  
to demonstrate the tremendous influence  
uch an advance in pure physics may have  
n the life of the community will be held  
the Science Museum, South Kensing-  
on, remaining open to the public for  
several weeks.

The National Committee for  
Mental Hygiene will meet at the Hotel  
Pennsylvania, New York City, November  
12-13. The annual program will be  
evoted to mental hygiene issues in  
Preparing for World Citizenship," as  
suggested by the preamble to the  
UNESCO constitution. The same theme  
has been chosen for the International  
Congress on Mental Hygiene to be held  
in London in August 1948. An examina-  
tion of the International Bill of Rights  
in relation to mental hygiene will be  
ollowed by a discussion of the mental  
health potentialities of the World Federa-  
tion for Mental Health, a voluntary  
agency, and the World Health Organiza-  
tion, a public agency of the UN for  
international cooperation. The annual  
luncheon meeting on November 13 will  
feature the presentation of the Lasker  
Award for the year's most significant  
contribution to popular adult education,  
especially in parent-child relationships,

and an address on the responsibilities  
and opportunities of citizens and mental  
hygiene organizations in the states in  
terms of immediate issues to be met.

## Elections

At the annual meeting of the Mt.  
Desert Island Biological Laboratory,  
Salsbury Cove, Maine, the following offi-  
cers of the Corporation were elected for  
1947-48: Dwight E. Minnich, University  
of Minnesota, president; Wm. H. Cole,  
Rutgers University, vice-president; Roy  
P. Forster, Dartmouth College, secre-  
tary; John Whitcomb, Bar Harbor, treas-  
urer; and Mrs. H. V. Neal, Salsbury  
Cove, clerk. Following the resignation of  
Roy Forster, who served 7 years as direc-  
tor, J. Wendell Burger, Trinity College,  
Hartford, Connecticut, was elected as the  
new director. In the reports of the officers  
it was noted that the Laboratory was  
having one of its most successful seasons.  
Thirty-seven workers and their assistants  
were in attendance.

The American Academy of Dental  
Medicine, at its first annual meeting in  
June, awarded an honorary membership  
to Walter Henry Wright, dean, New York  
University College of Dentistry. Officers  
of the Academy elected for the coming  
year are: Sidney Sorrin, president; J.  
Lewis Blass, president-elect; William M.  
Greenhut, secretary; George Bruns, trea-  
surer; and Allan N. Arvins, editor.

## NRC News

The Radio Corporation of America  
Fellowship Board of the NRC announces  
the first series of awards to five young sci-  
entists. These fellowships for the academic  
year 1947-48 provide for advanced gradu-  
ate study and research in the broad field  
of electronics. Recipients of the fellow-  
ships are: Arnold S. Epstein, B.S. in elec-  
trical engineering, Lehigh University, for  
graduate study at the University of Penn-  
sylvania with special reference to silenium  
and other rectifiers as variable capacitors;  
Willis W. Harman, B.S. in electrical en-  
gineering, University of Washington, for  
graduate study at Stanford University on  
the use of microwaves in certain cavity  
oscillators; Arnold R. Moore, B.S. in  
chemistry, Polytechnic Institute of  
Brooklyn, for graduate study at Cornell  
University on electronic properties of  
semiconductors; Sol Raboy, A.B. in phys-  
ics, Brooklyn College, for graduate study

at Carnegie Institute of Technology on  
the properties of semiconductors and their  
use as crystal counters; and H. Gunther  
Rudenberg, S.B. and M.A. in physics,  
Harvard University, for continuation of  
graduate study at Harvard University on  
the operation and design of wide-band  
pulse amplifiers.

These fellowships have been made pos-  
sible by a grant to the NRC from the  
Radio Corporation of America, Inc., for  
the purpose of increasing the number of  
trained scientific personnel and for the  
furtherance of electronics and closely re-  
lated fields. The selection of the fellows  
and the administration of the fellowship  
program are under the direction of the  
RCA Fellowship Board, the members of  
which are Frederick E. Terman, chair-  
man; C. C. Chambers, W. G. Dow,  
Frederick M. Feiker, R. Clifton Gibbs,  
I. I. Rabi, and Lloyd P. Smith.

Applications for awards for the academic  
year 1948-49 must be filed before Feb-  
ruary 1. Stipends will be from \$1,600 to  
\$2,100 a year, and an added amount, not  
to exceed \$600, may be provided annually  
to the institutions to which the fellow is  
assigned for tuition or necessary equip-  
ment.

Further information concerning these  
fellowships and application blanks may  
be secured from the Secretary of the RCA  
Fellowship Board, National Research  
Council, 2101 Constitution Avenue,  
Washington 25, D. C.

## Recent Deaths

John L. Ulrich, 69, physiologist and  
former faculty member at the Johns Hop-  
kins Medical School, the University of  
Maryland, Catholic University, and head  
of the Physiology Department, New York  
Homeopathic Medical College 1928-32,  
died in Baltimore on August 8.

Tiffany J. Williams, 49, professor of  
obstetrics and gynecology, University  
of Virginia, died August 19 in Charlotts-  
ville, Virginia.

Frederick M. Pederson, 78, formerly  
associate professor of mathematics, City  
College of New York, died August 20 at  
Hewletts Landing, Lake George, New  
York.

Willard L. Severinghaus, 65, a  
member of the physics staff at Columbia  
University from 1907 until his retirement  
in 1941, died August 28 in New York  
City after a long illness.

**William E. Wickenden**, 64, president of Case Institute of Technology since 1929, died of a heart attack at his summer home in Jaffrey, New Hampshire, on August 31, the day of his retirement from the presidency.

**Shuichi Kusaka**, 31, assistant professor of physics, Princeton University, was drowned at Beach Haven, New Jersey, on August 31.

**Rudolf W. Glaser**, 59, entomologist and zoologist who for the past 27 years had been associated with the Rockefeller Institute for Medical Research, died on September 4 in Princeton, New Jersey.

**John Herr Musser**, 64, professor of medicine at Tulane University since 1925, died September 5 in New Orleans.

**Robert G. Green**, 52, professor of bacteriology, University of Minnesota, since 1929, died September 6 at his home in Minneapolis.

**Frank O. Ellenwood**, 68, John Edson Sweet professor of engineering, Cornell University, and an authority on steam-power plants, died in Strong Memorial Hospital, Rochester, New York, on September 7.

**James S. Plant**, 57, director of the Essex County Juvenile Clinic, Newark, New Jersey, since 1923, died September 7 at his home in South Orange, New Jersey, following a heart attack.

**Rear-Admiral James C. Pryor**, 76, onetime head, Department of Hygiene, Naval Medical School, and professor of preventive medicine, George Washington University, died September 8 in the Naval Hospital, Brooklyn, after a long illness.

**Magel C. Wilder**, 49, assistant professor of biology, Brown University, died September 8 at her summer home in Hamilton, Rhode Island.

**Soup fin sharks and ling cod** will provide the raw materials for a new vitamin-concentrating plant now being built for Vitamin Sales Corporation of South Africa at Simonstown, about 23 miles from Cape Town. Oil from the livers of these fish contains high concentrations of Vitamin A. The technique called the Solexol Process, developed by the M. W. Kellogg Company, chemical engineers

of New York, will be employed. By this processing technique over 90 per cent of the original Vitamin A and D content of a crude fish oil is recovered in the concentrate. In its location close to abundant raw materials, its use of this process, and with a ready market for its various products, the Solexol vitamin unit, first of its kind in the world, is expected to set a pattern for future marine oil processing plants.

**The fall campaign of the New York University-Bellevue Medical Center Fund** will be officially inaugurated at a dinner on September 30 at the Waldorf-Astoria Hotel to be attended by over 1,500 leaders in medicine and industry. The principal speaker will be Frank W. Abrams, chairman of the Board, Standard Oil Company of New Jersey. In a discussion of "Medicine's New Role in Industry" Harry Woodburn Chase, chancellor, New York University, Robert Kehoe, director, Kettering Laboratory, Cincinnati, and Howard A. Rusk, chairman of the University's Department of Rehabilitation, will complete the list of speakers.

**Staticin Caronamide**, a new drug inhibiting the renal tubular excretion of penicillin and elevating the concentration of penicillin in plasma from two to seven times, has been reported in the August issue of *The American Journal of Medicine*. The clinical tests were conducted at the Pennsylvania Hospital, Philadelphia, by Christopher C. Shaw, William P. Boger, and J. William Crosson, Medical Research Division, Sharp & Dohme, Inc., and Walter W. Kemp, William S. M. Ling, and Garfield G. Duncan, Department of Medicine, The Pennsylvania Hospital. The results of the work have indicated that the most satisfactory administration of the new drug is by mouth every three hours. The Staticin-penicillin therapy is expected to be of greatest value in combating subacute bacterial endocarditis and osteomyelitis.

**The National Bureau of Standards** has established a new Division of Building Technology, the nucleus of which consists of the following sections: structural engineering; fire protection; heating, ventilating, and air-conditioning; exterior and interior coverings; and codes and specifications. Research will be limited to physics, chemistry, and engineering. Much of the work to be carried on will be primarily for the benefit of other governmental agencies, although

the results should be useful to the construction industry and it is intended that they shall be made available for purpose.

**Acta Chemica Scandinavica**, a journal in the field of chemistry, has been inaugurated by a group of Scandinavian scientists to further research done in various Scandinavian countries and as a means of presenting new ideas and results to colleagues all over the world. The journal aims thus also at a true international change of scientific information in the field of chemistry. All papers in the journal will be written in English, French, or German and by Scandinavian scientists or others working in Scandinavian laboratories. The journal will appear 10 times per year, with a total volume totaling about 1,200 pages. Subscription rate is \$8.50 plus postage. Orders should be addressed to Einar Munstergaard, Nørregade 6, Copenhagen, Denmark.

**According to an announcement by Science Service**, a second famous Harvard telescope has arrived in Torun, Poland, to become the chief instrument of the new observatory at the birthplace of Nicholas Copernicus. The 8-inch Draper telescope, which provided spectra of 100,000 northern stars for the late Annie Jump Cannon's 10-volume Draper catalogue of spectral classifications, and brightness, has been especially rehabilitated and equipped for a Polish program of research on stellar colors, temperatures, and chemical compositions.

#### Make Plans for—

**American Institute of Electrical Engineers**, Middle Eastern District Meeting, September 23-25, Dayton, Ohio.

**American Public Health Association**, October 6-10, Atlantic City, New Jersey.

**American Academy of Ophthalmology and Otolaryngology**, October 12-14, Chicago.

**Society of Rheology**, October 30-31, New York City.

**American Association for the Advancement of Science**, 114th Meeting, December 26-31, Chicago, Illinois.



# COMMENTS

## by Readers

**Promotion and inhibition of growth** may be produced by the same substance at different concentrations. Thimann (*Amer. J. Bot.*, 1937, 24, 407-412; quoted from Meyer and Anderson. *Plant physiology*. New York: D. Van Nostrand, 1939. P. 580) suggests that roots, buds, and stems all react in a comparable way to auxin, their growth being inhibited by relatively high and promoted by relatively low auxin concentrations, (and that this) is a possible explanation of the contrasting effects of auxins upon elongation in roots and aerial organs. Elongation of roots is favored only at very low concentrations; at all higher concentrations their growth is checked. Stems and coleoptiles show a similar behavior except that the optimum range of concentrations for elongation is much higher than for roots. The same concentrations of auxins which favor stem elongation result in a retardation of root elongation. . . . Briefly, therefore, whether auxin will exert an accelerating or an inhibiting effect upon growth seems to depend in part upon its concentration and in part upon the specific tissue involved."

Engard and Nakata (*Science*, May 30, pp. 577-580) present evidence for the presence in sugar cane of both a growth inhibitor and a growth promotor. In view of the above findings by Thimann and the more recent work by Eyster (*Plant Physiol.*, 1946, 21, 68-74) it seems very likely that the growth inhibitor and growth promotor in sugar cane are one and the same substance, producing differences in growth reactions because of differences in concentration. It is expected that acid will extract the growth substance more completely and more rapidly than water. The "acid fraction" may also owe some, or a dominant part, of its activity to the tartaric acid which was used for extraction purposes.

For comparative reasons it would be well to have a control using tartaric acid without any extract and another control using water without any extract.

It would also be well to determine the *Avena* curvature produced by dilutions of the acid fraction. Dilutions of 10X, 100X, 1,000X, and 10,000X are suggested. Controls of these dilutions should also be run, and curvature determinations should be made. When this is done, there is a reasonable feeling of confidence that these isolated bits of evidence on auxins can be brought into agreement and perhaps result in a somewhat more complete picture of their true activity. (H. C. EYSTER, *Charles F. Kettering Foundation*, Yellow Springs, Ohio.)

**Description of centrifuge operations in terms of duration and speed** is indeed inadequate, as Costello points out (*Science*, May 2, p. 474). It is unfortunate that correspondence directed toward correction of an incomplete system of description should contain a looseness of terminology which beclouds the issue.

The formula,  $c' = \frac{4\pi^2 n^2 r}{g}$ , attributed by Costello to Heilbrunn, has been a part of the ordinary college course in general physics for decades,  $c'$  representing not "the centrifugal force in terms of gravity" but the ratio of the central acceleration (accompanying uniform circular motion) to the standard value of the acceleration due to gravity. More concisely,  $c'$  is the "centripetal acceleration in g's." It is not a force. ( $n$  is the number of revolutions per second;  $g$ , the standard acceleration due to gravity, in cm./sec.<sup>2</sup>; and  $r$ , the distance, in centimeters, from the center of rotation to the material.)

The distinction between acceleration and force is far from trivial in centrifuging, for the separation of materials of different densities results from the difference in force. However, at a given speed and given radius from the axis of rotation, the acceleration is the same for all materials; the separation is produced by unbalance of forces arising from non-uniformity of density. It is therefore appropriate to describe the centrifuging

in terms of  $c'$ , but this is *centripetal acceleration*, not centrifugal force.

The emphasis given by Costello to the importance of estimating  $r$  with great care should be balanced by the remark that the percentage uncertainty in  $r$  may reasonably be twice that in  $n$ , since  $r$  occurs in the first power, while  $n$  occurs in the second power (Eshbach's *Handbook of engineering fundamentals*. New York: John Wiley, 1936). In this connection there would be considerable value in following the practice of specifying the percentage uncertainty in  $r$  and  $n$  in reporting centrifuge operations.

Finally, the formula should be reduced for convenience to the form

$$c' = 4.029 \times 10^{-2} n^2 r,$$

which is obtained by substituting numerical values for  $4\pi^2$  and for the standard value of  $g$ . (HOMER C. KNAUSS, *Raytheon Manufacturing Company*, Waltham, Massachusetts.)

In regard to the results of creatinine, hippurate, and inulin clearance estimations in rats by M. Friedman, Dicker and Heller (*Science*, August 8, p. 127) point out the possible inhibition by pain and anesthesia of urine flow in rats. We have recently conducted experiments on humans employing prolonged exposure (15 minutes) of one limb in an ice-water bath. Our results show a pronounced inhibition of urine excretion during, but particularly following, this experience. This phenomenon was associated with an increase in specific gravity of the urine and, in some instances, a change in protein excretion per minute. Experiments conducted under anesthesia (ethylene, cyclopropane, and various levels of spinal anesthesia) indicate that this pain stimulates the liberation of an antidiuretic hormone. The site of liberation is probably not within the immersed limb or within the kidney (since it was not present in high levels of spinal anesthesia if the lower extremity was used). Evidence supports its origin as being above the level of cervical 4. It was also our impression that the site of liberation was the posterior pituitary gland, although we were unable to show conclusively by bioassay on rats that the serum concentration of antidiuretic hormone increased in the patients so tested. (L. D. ODELL and G. T. ARAGON, *Department of Obstetrics and Gynecology*, University of Chicago.)

## Relative Sensitivity of Dormant and Germinating Seeds to 2,4-D

JOHN W. MITCHELL and JAMES W. BROWN<sup>1</sup>

Bureau of Plant Industry, Soils, and Agricultural Engineering,  
Beltsville, Maryland

It has been reported that seeds or seedlings can be killed by adding 2,4-D to soil or manure in which they occur (1-4). Later experiments at the U. S. Plant Industry Station have shown, however, that some seeds will eventually germinate, although the manure or soil in which they occur is treated with sufficient 2,4-D to be lethal for a period of several weeks. These results suggest that seeds of some species may be relatively insensitive to 2,4-D while dormant, and that those remaining dormant in soil or manure until the chemical is inactivated, may not be affected. Experiments described here were made to study the effect of treating dormant and germinating seeds with 2,4-D.

In the first experiment mustard seeds (variety Southern Giant Curled) were incubated on moist blotting paper in

TABLE 1  
RELATIVE ROOT ELONGATION OF MUSTARD SEEDLINGS TREATED FOR 30 SECONDS WITH CONCENTRATIONS OF THE AMMONIUM SALT OF 2,4-D AT 4 STAGES OF DEVELOPMENT\*

Concentrations (ppm)	Stage of development at treatment			
	I	II	III	IV
0	100	100	100	100
10				76
100	120	72	46	35
500	87	25	0	0
1,000	31	9	0	
2,000	12	7	0	

\* Values represent percentages of elongation of radicles, the respective controls rated as 100 per cent.

Petri dishes at 25°-27°C. until they had reached the desired stages of germination. Treatments were applied after the seeds had reached the following stages: I, unswollen seeds immediately after wetting; II, swollen seeds; III, seeds that had swollen and ruptured the coats; IV, seeds that had developed radicles 5 mm. in length. In treating, selected seeds or seedlings were immersed in the desired concentrations of aqueous solutions of the ammonium salt of 2,4-D (ammonium 2,4-dichlorophenoxyacetate) for definite periods of time. They were immediately washed for 15 seconds with running distilled water, blotted, and incubated. Untreated seeds were immersed for comparable periods in distilled water and designated controls. After both the controls and those treated with the salt had reached stage III, elongation of the radicles was recorded for the following 24-hour period. In the case of stage

<sup>1</sup> The authors wish to acknowledge the assistance given by E. H. Toole and Vivian Toole which made this study possible.

IV, elongation of the radicles was measured for a 24-hour period immediately after they had reached a length of 5 mm.

Root growth of mustard seeds treated at stage I for 10 minutes in a solution containing 500 ppm of the ammonium salt of 2,4-D was reduced 70 per cent. A 1-second treatment of seeds in stage IV completely checked radicle elongation.

The sensitivity of mustard seeds was also determined by treating selected seeds in different stages of germination with a solution containing 500 ppm of the salt for a period of 30 seconds. On the basis of subsequent root growth, resting seeds were relatively insensitive to the salt. The seeds became more sensitive as they absorbed water and ruptured their coats. Seeds from which the radicles protruded were extremely sensitive to the salt solution (Table 1, Fig. 1).

Seeds of subterranean clover (5) (*Trifolium subterraneum*) germinate readily when subjected to a temperature of 20°C., on the other hand, they absorb water, yet remain dormant when subjected to a temperature of 30°C. It is thus possible to plant the seeds in treated soil and hold them dormant for the period during which the acid remains active in soil (4). Once

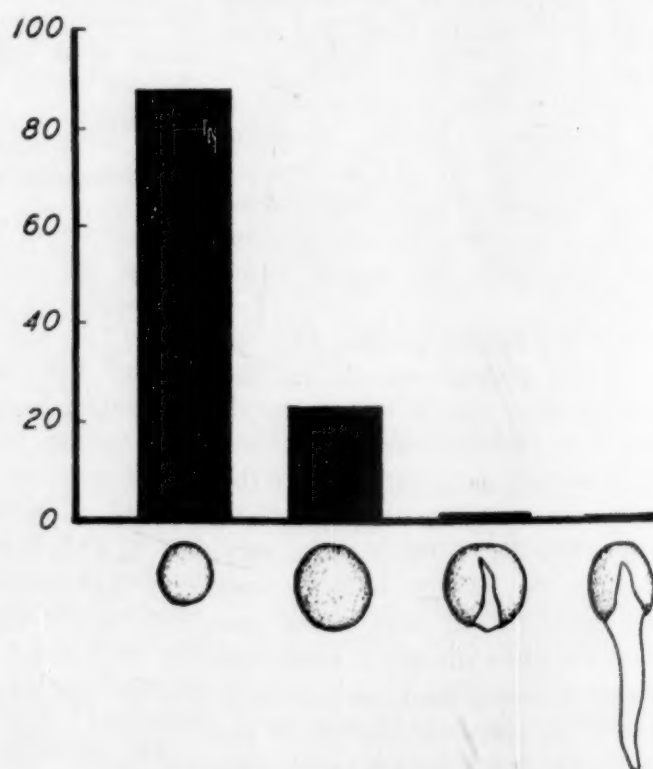


FIG. 1. Effect of ammonium 2,4-dichlorophenoxyacetate on root growth of mustard seedlings treated at the four stages of germination indicated by the drawings. The bars represent percentages of root elongation for the four treated lots, the respective controls being considered as 100 per cent.

the 2,4-D has become inactivated, the seeds and soil can then be transferred to a temperature favorable for germination and the effect of the previous chemical treatment observed.

The clover seeds were first placed on moist blotters at controlled temperature of 30°C., a condition under which they would absorb water but remain dormant with the seed coats intact. At intervals, samples of the seeds were weighed to



determine when the maximum amount of water had been absorbed. They were then divided into 20 groups of 25 selected seeds each. Five groups were planted individually in 0.45 pound of moist, composted soil contained in individual waxed-paper boxes. Five other groups were planted in individual boxes containing an equal amount of composted soil with which 2,4-D had been mixed at the rate of 4 pounds/acre. All 10 boxes were placed at a controlled temperature of 20°C. The same two procedures were then repeated using the remaining groups of seed, but these were maintained in the moist soil at a constant temperature of 30°C.

Seeds in the untreated soil at 20°C. germinated and grew vigorously within a few days after they were planted. Emergence and growth of those in the treated soil were greatly inhibited by the acid (Table 2). After 11 days at 20°C., some plants in the treated soil had grown above the surface, but they were deformed and stunted.

The seeds at 30°C. remained dormant. Inactivation of 2,4-D in the treated soil was determined by repeatedly planting mustard seeds in it. After 19 days, the 2,4-D was completely inactivated, as indicated by the fact that mustard seeds germinated, and the plants grew as vigorously in the treated as in the untreated soil. Boxes containing the treated and those containing the untreated soil were then transferred to 20°C. The clover seeds in both germinated and grew vigorously, none being deformed (Table 2).

It is evident from results with this species of clover that the seeds were relatively resistant to 2,4-D while dormant, but

## Inactivation of Certain Neurotropic Viruses *in Vitro* by Serum Lipids

J. CASALS and PETER K. OLITSKY

The Rockefeller Institute for Medical Research,  
New York City

The inactivation of viruses by some lipids, particularly fatty acids and their soaps, is well known (1) and has been attributed generally to the surface action of the chemicals. While investigating various aspects of the neutralization test with neurotropic viruses, a slight but definite and reproducible reduction of the titer of the viruses was noted when the latter were incubated with clear sera deriving from several normal animal species, as compared with that shown by viruses in buffered saline solution controls. Moreover, the lipid fraction obtained from such sera either by acetone-ether or by hot alcohol extractions, emulsified in buffered saline solution and added to a virus suspension, induced rapid and pronounced reduction in titer—a drop of .01 to .0001 within 5 minutes.

The test was carried out by mixing equal volumes of a 1:500 suspension of infected mouse-brain in buffered saline solution and of the emulsion of lipid; as a control, a similar mixture was used in which buffered saline solution substituted for the emulsion of lipid. The pH was maintained at 7.0–7.2. The mixtures were then placed in a water bath at 37°C., portions being removed at intervals from 5 minutes to 24 hours and titrated in mice by means of intracerebral injection. Serum lipids from

TABLE 2

PERCENTAGES OF EMERGENCE OF SUBTERRANEAN CLOVER (*Trifolium subterraneum*) IN UNTREATED SOIL (U) AND IN SOIL TO WHICH 2,4-D WAS ADDED IN AMOUNTS EQUIVALENT TO 4 POUNDS/ACRE (T)

Replications	7 days at 20°C.		11 days at 20°C.		19 days at 30°C.; 3 days at 20°C.		19 days at 30°C.; 9 days at 20°C.	
	U	T	U	T	U	T	U	T
1	76	0	100	48	88	68	88	80
2	72	0	100	32	44	76	80	88
3	80	0	92	48	48	68	88	80
4	64	0	92	48	44	24	96	96
5	64	0	100	32	92	84	96	80
Average	71	0	97	42	63	64	90	85

became extremely sensitive to the acid after the seed coats were broken and the seeds began to germinate. Mustard seeds likewise proved to be relatively insensitive to 2,4-D when the compound was applied to the resting seed. Mustard seeds in later stages of germination were extremely sensitive to the compound. It is indicated on the basis of these results that 2,4-D is likely to be most efficient as a preplanting soil treatment for killing seeds of obnoxious plants if applied at a time when the greatest number of such seeds are germinating.

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TABLE 1

INTRACEREBRAL TITER OF JAPANESE B ENCEPHALITIS VIRUS FOLLOWING INCUBATION AT 37°C. IN THE PRESENCE OF LIPIDS

Test	Virus suspended in:	Length of incubation			
		5 min.	4 hr.	8 hr.	24 hr.
1	Buffered saline (control)	10 <sup>-8.8</sup>	10 <sup>-8.2</sup>	10 <sup>-8.5</sup>	10 <sup>-8.6</sup>
	Horse serum lipids	10 <sup>-5.8</sup>	10 <sup>-3.3</sup>	<10 <sup>-2.5</sup>	<10 <sup>-2.6</sup>
2	Buffered saline (control)	10 <sup>-8.7</sup>		10 <sup>-8.5</sup>	10 <sup>-7.2</sup>
	Egg-yolk lipids	10 <sup>-8.3</sup>		10 <sup>-9.3</sup>	10 <sup>-6.8</sup>
	Human brain lipids	10 <sup>-7.6</sup>		10 <sup>-6.6</sup>	10 <sup>-6.6</sup>

mice, hamsters, rabbits, and horses, have been found active; for practical reasons horse serum has been used extensively. The viruses thus far tested with similar results were those of St. Louis, Russian Far East, and Japanese B encephalitis. Test 1 (Table 1) shows a typical experiment.

The viral inactivation took place in the presence of serum protein at 2–4°C., although more slowly, and the inactivating agent was not dialyzable and withstood heating at 99°C. for 1 hour. There was no correlation between the change in surface or interfacial tension brought about by the lipid and the degree of viral inactivation.

Fractionation of serum lipids by using several solvents showed that when serum was extracted in succession with acetone, ether, and hot ethyl alcohol-ether, each extract was effective in inactivating virus. Lipids from other animal sources have been tested. Whole lipids obtained from egg yolk were not effective even after 24-hour incubation; those deriving from brain showed only a moderate activity (Test 2). On frac-

tionation of the brain lipids, the acetone fraction had a marked inactivating action as well as a low surface tension; the ether and hot alcohol-ether fractions were only moderately active, irrespective of surface tension.

The inactivation of neurotropic viruses by serum lipids is now being investigated in connection with (a) the mechanism, including influence of the degree of dispersion, and closer identification of the active agent; (b) the bearing of the results on the neutralization test as it is now carried out; and (c) its possibilities as the basis for chemotherapy of experimental infections.

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## The Abundance of Thallium in the Earth's Crust

L. H. AHRENS

*Department of Geology,  
Massachusetts Institute of Technology*

The most generally accepted value for the abundance of thallium in the earth's crust is that given by Goldschmidt (2) as 0.00003 per cent Tl, by weight.

Although analytical data on the abundance of thallium in rocks are very meager (most analyses for thallium are confined to pegmatitic minerals), it should be possible to make a reasonably accurate estimate of its abundance in the earth's crust as a result of its close association with rubidium in minerals. It has been shown (1) that, with the exception of some sulfide minerals where thallium alone may be present, rubidium and thallium are confined essentially to potassium minerals and the cesium mineral, pollucite, and that in these the ratio Rb/Tl varies relatively slightly from area to area and is independent of the type of potassium host mineral (mean weight ratio per cent Rb/per cent Tl = 100). A plot of log per cent  $Rb_2O$  vs. log per cent  $Tl_2O$  for about 170 mineral specimens so far investigated, covering a thousand-fold concentration range, shows that throughout this range a straight line of unit slope accommodates the plotted points most satisfactorily. Consequently, one may infer that in the earth's crust as a whole the ratio Rb/Tl may be regarded as equal to about 100.

The value of the abundance of rubidium (0.03 per cent Rb) given by Goldschmidt (2) is probably reasonably accurate, although this value might be altered slightly because of several relatively recent rubidium determinations on rocks; hence the abundance of thallium in the earth's crust is  $0.03/100 = 0.0003$  per cent. One may infer, therefore, that the older value (0.00003 per cent) is low by a factor which may be as high as about 10.

It may be noted that, as mentioned above, thallium has a dual geochemical behavior and is usually present as a trace in many sulfides. In comparison with the quantity of thallium in the vast amounts of potash minerals, however, the thallium contained in relatively small quantities of sulfide minerals is probably insignificant; if significant, its presence in sulfide

minerals would tend to increase the factor of 10 referred to above.

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## Reaction of Certain Plant Growth Regulators With Ion Exchangers

ROBERT J. WEAVER

*Department of Botany, University of Chicago*

Several investigators have reported that 2,4-D and certain other plant growth regulators may be readily leached from some soils (2, 3, 5). The fact that large volumes of water did not always completely remove the 2,4-D would indicate that some of the compound may be adsorbed by the soil. It was recently shown that 2,4-D is inactivated by adsorption on charcoal (4).

The degree of adsorption of several plant growth regulators by certain ion exchange materials and the readiness with which the compounds are eluted after having been adsorbed are reported here.

The cation exchangers used were a resin exchanger, Amberlite IR-100,<sup>1</sup> a carbonaceous exchanger, Zeo-Karb H, a synthetic sodium aluminosilicate, Decalso, and a processed glauconite, Zeo-Dur. The anion exchangers were the amine resins, Amberlite IR-4B and De-Acidite.

All cation exchangers were screened -20 + 40 mesh. IR-100 and Zeo-Karb H were treated with a number of portions of 5 per cent hydrochloric acid, with frequent stirring, for a period of 48 hours; Decalso and Zeo-Dur, with several changes of 4 per cent sodium or calcium chloride over a period of 24 hours. After the materials were thoroughly washed with distilled water they were dried in an oven at 40°C. The anion exchange materials were prepared by stirring the samples with portions of 5 per cent sodium carbonate over a 24-hour period, washing, and then drying for about 5 hours at 40°C.

Six plant growth regulators were used for experimentation: 2,4-dichlorophenoxyacetic acid (2,4-D), ammonium 2,4-dichlorophenoxyacetate ( $NH_4 2,4-D$ ), cupric 2,4-dichlorophenoxyacetate [ $Cu(2,4-D)_2$ ], calcium 2,4-dichlorophenoxyacetate [ $Ca(2,4-D)_2$ ], 2,4,5-trichlorophenoxyacetate (2,4,5-T), and isopropyl N-phenylcarbamate (IPPC). The 2,4-D was purified by running it through several salt-acid cycles.  $Cu(2,4-D)_2$  was prepared by reacting an excess of an aqueous solution of  $NH_4 2,4-D$  with a solution of cupric chloride, and then washing the precipitate of  $Cu(2,4-D)_2$  free of ammonium chloride.  $Ca(2,4-D)_2$  was made by adding calcium chloride to an aqueous solution of  $NH_4 2,4-D$ . The spectrophotometric method developed by Bandurski (1) was employed to measure the compounds in solution.

Static trials were divided into two types: (1) those in which it was determined how much of a compound an exchanger material removed from a solution, and (2) those in which elution of regulators from exchangers was studied.

In static trials 0.5-gram samples of the exchanger were placed in 250-ml. Erlenmeyer flasks. Fifty ml. of the appro-

<sup>1</sup> The Amberlites were obtained from the Resinous Products and Chemical Company and other exchangers from the Permutit Company.



appropriate solutions were then pipetted into the flasks, which were then stoppered and allowed to stand for 48 hours, with frequent shaking. At this time the concentrations of the growth regulator in the supernatant liquid were determined, and the amounts of compound adsorbed or eluted calculated.

In elution studies the regulators were dissolved in 95 per cent ethyl alcohol and shaken with the exchangers for about 30 minutes. The alcohol was then evaporated by placing the container in a circulating oven at about 70°C. The growth regulators were also added to exchanger materials by shaking the exchangers with aqueous solutions of the compounds. The amount of regulator adsorbed by the exchanger was calculated by measuring the decrease in concentration of the compound in the supernatant liquid.

however, adsorbed less  $\text{Cu}(2,4\text{-D})_2$  than 2,4-D,  $\text{Ca}(2,4\text{-D})_2$ , or  $\text{NH}_42,4\text{-D}$ . These results indicate that when 2,4-D or its salts are added to soils in chemically equivalent quantities, equal amounts of the anion might be adsorbed by the soil. This might not hold true in soils containing anion or acid exchanging materials. IR-100 H adsorbed less 2,4,5-T than 2,4-D or its salts, but adsorbed much more IPPC. With the anion exchanger the situation was reversed.

It was determined that IPPC was strongly adsorbed by IR-100 in the hydrogen, sodium, or calcium cycle, while 2,4-D was strongly adsorbed only by the resin in the hydrogen cycle. Much less 2,4-D was adsorbed by Amberlite IR-100 H when the pH of the solution was 3.3 than when it was 2.5 or lower. Perhaps acidic soils would have higher adsorptive capacities

TABLE 1  
ADSORPTION OF PLANT GROWTH REGULATORS BY EXCHANGER MATERIALS\*

Compound	Exchanger														
	IR-100 H			Zeo-Karb H			Decalso			Zeo-Dur			De-Acidite		
	Micro eq. adsorbed	Mg. adsorbed, 1/4 gram oven-dry exchanger	% adsorbed	Micro eq. adsorbed	Mg. adsorbed, 1/4 gram oven-dry exchanger	% adsorbed	Micro eq. adsorbed	Mg. adsorbed, 1/4 gram oven-dry exchanger	% adsorbed	Micro eq. adsorbed	Mg. adsorbed, 1/4 gram oven-dry exchanger	% adsorbed	Micro eq. adsorbed	Mg. adsorbed, 1/4 gram oven-dry exchanger	% adsorbed
2,4-D	5.78	1.46	51.1	11.0	2.58	97.3	00	00	00	00	00	00	5.23	1.83	46.2
$\text{NH}_42,4\text{-D}$	5.97	1.62	52.8	11.0	2.78	97.3	00	00	00	.16	.041	1.41	5.08	1.91	44.9
$\text{Cu}(2,4\text{-D})_2$	5.72	1.64	50.6	11.0	2.93	97.3	00	00	00	.01	.003	0.09	3.42	1.36	30.2
$\text{Ca}(2,4\text{-D})_2$	6.13	1.68	54.2	11.1	2.82	98.1	.27	.70	2.39	.18	.047	1.59	5.49	2.08	48.5
2,4,5-T	4.77	1.39	42.2	11.0	2.98	97.3	00	00	00	.73	.201	6.45	6.17	2.49	54.6
IPPC	8.70	1.78	76.9	11.2	2.13	99.0	.45	.87	3.98	.43	.083	3.80	1.03	0.29	9.1

\* Each figure is average of 3 replicates.

Pyrex glass tubes about 40 mm. in diameter and 25 cm. in height were used for studies of leaching. The tubes were tapered at the lower end so that the lower 5 cm. was about 1/4 inch in diameter. The exchanger beds, which were about 10 cm. in height, were supported by a plug of glass wool at the point of constriction of the tube. A reservoir of liquid could be maintained above the exchanger material. A piece of rubber tubing about 1 1/2 inches long and with a screw-type clamp was fitted over the lower end of the leaching tube.

After the exchanger material was placed in the tube, distilled water was slowly forced into the lower end until the level of water reached the top of the exchanger bed, after which the screw clamp was closed. This backwashing was necessary in order to prevent presence of air pockets in the bed. After the bed was allowed to stand for 15 minutes, 55 ml. of distilled water was added above it. The screw clamp was then opened so that a slow drip rate was obtained which allowed 55 ml. of water to leach through in about 45 minutes.

The relative adsorptive capacities of 6 exchanger materials for 6 plant growth regulators were studied. Aqueous solutions of 0.000226 normality of 2,4-D (50 ppm),  $\text{NH}_42,4\text{-D}$ ,  $\text{Cu}(2,4\text{-D})_2$ ,  $\text{Ca}(2,4\text{-D})_2$ , 2,4,5-T, and IPPC were prepared. IPPC was considered to have a valence of 1. Fifty ml. of these solutions was added to 0.439-gram samples of IR-4B or 0.5-gram samples of all other exchangers. The data in Table 1 indicate that IR-100 H and Zeo-Karb H adsorbed much of the growth regulators, while the Decalso and Zeo-Dur adsorbed little or none. IR-100 H adsorbed almost equal micro equivalents of 2,4-D and its salts. De-Acidite and IR-4B,

for 2,4-D than alkaline soils. The amount of 2,4-D adsorbed by ion exchangers from solutions varying in concentration from 20 to 200 ppm was about directly proportional to the concentration of the 2,4-D in the initial solution.

Less elution of 2,4-D from Amberlite IR-100 H was caused by 0.75 N solutions of NaCl,  $\text{CaCl}_2$ , or  $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$  than by

TABLE 2  
ELUTION OF 2,4-D FROM 0.5-GRAM SAMPLES OF IR-100 H BY WATER AND 0.75 N SOLUTIONS CONTAINING A MONO-, DI-, OR TRIVALENT CATION

Solution	2,4-D eluted	2,4-D eluted
	(mg.)	(%)
water	0.685	45.7
HCl	0.720	48.0
NaCl	0.335	22.3
$\text{CaCl}_2$	0.370	24.7
$\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$	0.436	29.1

water (Table 2). Similar results were obtained with solutions containing anions of three different valences. IPPC, however, was eluted from IR-100 H by NaCl solution. Hydrochloric acid readily eluted 2,4-D from the acid exchangers IR-4B and De-Acidite. These data suggest that much of the 2,4-D in soil remains adsorbed and not subject to leaching.

Comparative rates of leaching of 2,4-D,  $\text{NH}_42,4\text{-D}$ ,  $\text{Cu}(2,4\text{-D})_2$ , 2,4,5-T, and IPPC from beds of Decalso and Zeo-Dur about 10 cm. in height were studied. One-half or more of all the compounds were usually leached from the exchanger

materials by 55 ml. of water, and the compounds were usually removed in about the same amounts. The relatively soluble  $\text{NH}_4\text{2,4-D}$  was not removed in greater quantities than 2,4-D, which is of much lower solubility. It seems probable that in a soil 2,4-D or a relatively insoluble 2,4-D salt might be converted to soluble forms by ammonium, sodium, or other ions present in the soil solution.

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## Granulosa Cell Tumors in Intrapancreatic Ovarian Grafts in Castrated Mice<sup>1</sup>

MIN HSIN LI and W. U. GARDNER

*Department of Anatomy,  
Yale University School of Medicine*

Biskind and Biskind (1) reported that granulosa cell tumors have developed in ovaries transplanted into spleens of three castrated female rats. Our previous experiments showed the formation of granulosa cell tumors and luteomas in intrasplenic ovarian grafts in castrated male and female mice (3). These studies were based on two principles: (1) the capability of the liver to inactivate ovarian hormones when the hormones circulate through the hepatic portal system, and (2) the increase of pituitary gonadotropins subsequent to castration. It was assumed that the prolonged stimulation, by augmented amounts of gonadotropic hormones, of intrasplenic ovarian grafts was responsible for the neoplastic growths. More recent investigations (2) revealed that the development of ovarian tumors in intrasplenic ovarian grafts was inhibited by administration of estradiol benzoate and testosterone propionate. The malignancy of the induced granulosa cell tumors was indicated by the ability to metastasize and to transplant in new hosts. The present experiment, using intrapancreatic ovarian transplantation in castrated mice, demonstrates that splenic tissues do not play a direct role in the pathogenesis of ovarian tumors arising in the grafts.

Male and female mice of A, C<sub>3</sub>H, CBA, and C<sub>57</sub> strains and hybrid mice were used. These were castrated and received, at the same time, an autoplasmic or homoplasmic ovarian graft in the pancreas. Among the first group of 5 experimental animals, two granulosa cell tumors and one pretumorous growth were found 168 days after grafting. No tumor was noted in two grafts with vascularized adhesions that permitted drainage through other than the hepatic portal system. One tumorous graft in a male mouse (C<sub>3</sub>H strain) was 7 x 8 x 10 mm. in diameter; the other, which developed in a female hybrid mouse (AC<sub>3</sub>), measured 10 x 11 x 13 mm. in diameter. The uterus of the latter animal weighed 75 mg. at autopsy. The pretumorous graft occurred in a male mouse of the A strain.

Microscopically, the granulosa tumor cells were arranged in a folliculoid pattern showing numerous mitotic figures.

<sup>1</sup> This research has been aided by grants from the Anna Fuller Fund and the Jane Coffin Childs Memorial Fund for Medical Research.

Some of the folliculoid structures contained hemorrhagic cavities. Luteinized cells and small necrotic areas were present, and a spicule of bone was observed at the periphery of one tumorous ovarian graft. Major portions of the tumors were separated from the pancreatic tissue by bursa-like spaces lined by germinal epithelium. No metastasis was observed in the liver. The pretumorous graft showed masses of tubular ingrowths from the germinal epithelium, and the transformation of some of the epithelial cells into granulosa tumor cells was noted. Thus, the morphology of granulosa cell tumors induced in the pancreatic site resembled that of the tumors developed in intrasplenic ovarian grafts. The present experiments are interpreted to substantiate further the assumption that overaction of gonadotropic hormones is responsible for the development of the ovarian tumors in mice.

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## Differential Phytotoxicity of Metabolic By-Products of *Helminthosporium victoriae*

FRANCES MEEHAN

*Botany and Plant Pathology Section,  
Iowa Agricultural Experiment Station*

H. C. MURPHY

*Bureau of Plant Industry, Soils, and Agricultural  
Engineering, U. S. Department of Agriculture*

The "*Helminthosporium* blight" of oats caused by *H. victoriae* Meehan and Murphy has developed so rapidly that it has attained the proportions of a major plant disease within two years after its discovery. Susceptibility is apparently limited to oat varieties and selections that possess the "Victoria-type" resistance to crown rust (*Puccinia coronata avenae* (Corda) Eriks. & E. Henn.). The unusually fast build-up of the disease has been facilitated by the widespread planting of large acreages to susceptible varieties.

The means by which *H. victoriae* causes necrosis has been the subject of some speculation. In a previous article (2) the suggestion was made that the pathogenic action of this fungus involves the production of a toxic substance. Inoculation tests with sterilized mycelium and filtered extracts from cultures have given evidence that a very potent toxin is secreted by the fungus, which is responsible for the characteristic longitudinal foliar striping or discoloration. Data regarding its production and effects are briefly summarized in this paper.

Evidently the basal infection of the oat plant is the only direct manifestation of parasitic action by *H. victoriae*, since the organism has not been isolated from the blighted leaves until after complete necrosis of the tissue. It may be that this

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fungus is too weak a parasite to establish infection in healthy tissue even of susceptible oat varieties without the help of toxic secretion in advance. Otherwise, it would be expected that a fungus that grows vigorously in culture would progress rapidly in the plant from the basal portions to the leaves.

Preliminary tests showed that the toxic substance or substances were readily formed in cultures grown on media containing either organic or inorganic nitrogen. In a typical experiment, cultures of *H. victoriae* were grown for 30 days at room temperature (24–28° C.) in flasks, each containing 100 ml. of Richard's solution, filtered through a Büchner funnel to remove the hyphal mass, and the filtrate then passed twice through a Berkefeld filter to render it aseptic. Boone and Clinton oat seedlings were grown for one week in nutrient

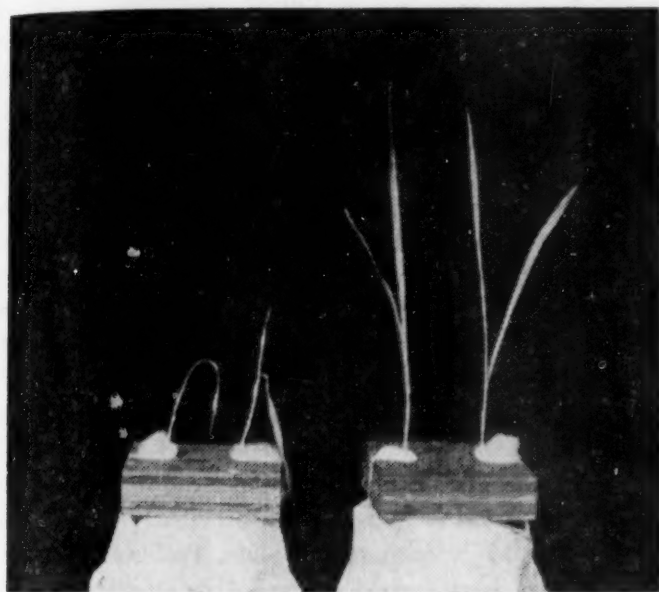


FIG. 1. Oat plants of varieties Boone (left) and Clinton (right) grown in nutrient solution, 90 hours after toxic extract, 1 part in 45 parts of water, had been added.

water culture, after which time the nutrient solution was replaced by the filtrate in a series of dilutions ranging from 1:15 to 1:1,800 in tap water. Observations made at 4-hour intervals showed the following reactions in the susceptible Boone variety: at dilutions of 1:90 or less the leaf blades became rigid and inflexible within 40 hours, and after 48 hours these leaves showed a slight twisting. A more critical indication of phytotoxicity was obtained at dilutions of 1:45 or less: the healthy green color of normal leaves changed to a dull grayish-brown after 52 hours. This color change preceded the death and drying of the leaves. The seedlings of the resistant variety, Clinton, were unaffected by the filtrate in these dilutions. Since it was found that Richard's solution alone, minus the amount of sugar equivalent to that used by the fungus in growth, was harmless to the susceptible plants, it may be concluded that a substance was formed as a metabolic by-product of the growth of *H. victoriae* that was toxic to the susceptible variety of oats. The color reaction is considered most reliable for bioassay technique, since it was obtained consistently at the same dilution range in a series of tests.

The toxin occurs in the cells of *H. victoriae* as well as in the nutrient medium, as shown by the fact that leaves of susceptible oat varieties were killed when sprayed with a water suspension of sterilized blended mycelium containing no culture substrate.

The toxic principle in the culture extract is relatively stable, as it was not destroyed by autoclaving for 20 minutes at 15 pounds pressure. Lee's work (1) with *H. sacchari* (Breda de Haan) Butler showed this fungus to have a strong capacity for reducing inorganic nitrates to nitrites which were assumed to be responsible for the toxicity of this organism to sugar-cane leaves. The production of toxin by *H. victoriae* on media containing only organic sources of nitrogen is evidence that nitrite formation is not the cause of this toxic action.

Some tapering-spored species of *Helminthosporium* have been found to produce characteristic intracellular chemical compounds (3) of the polyhydroxyxanthone series, such as *ravenelin* (3 methyl-1,4,8-trihydroxyxanthone) in *H. ravenelii* Curt. Further studies will be required to determine whether comparable materials are present in the toxic solutions from *H. victoriae*.

*H. victoriae* is primarily a facultative soil- and seed-borne saprophyte that possesses a low order of phytopathogenicity. It causes severe leaf blight in addition to basal stem and root necrosis without invading the plants extensively. The limited progress it does make may depend largely upon direct injury to plant tissues by the toxin. Varieties of oats such as Clinton, that are highly resistant to, or immune from, attack by the fungus itself, are likewise not injured by the toxic secretions. The extreme susceptibility to the fungus exhibited by the Victoria derivatives might be reasonably expected, since the resistance shown by these varieties to crown rust is dependent upon a hypersensitive reaction to the rust fungus, an obligate parasite.

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## Application of "Metabolite Antagonism" to Cancer Research

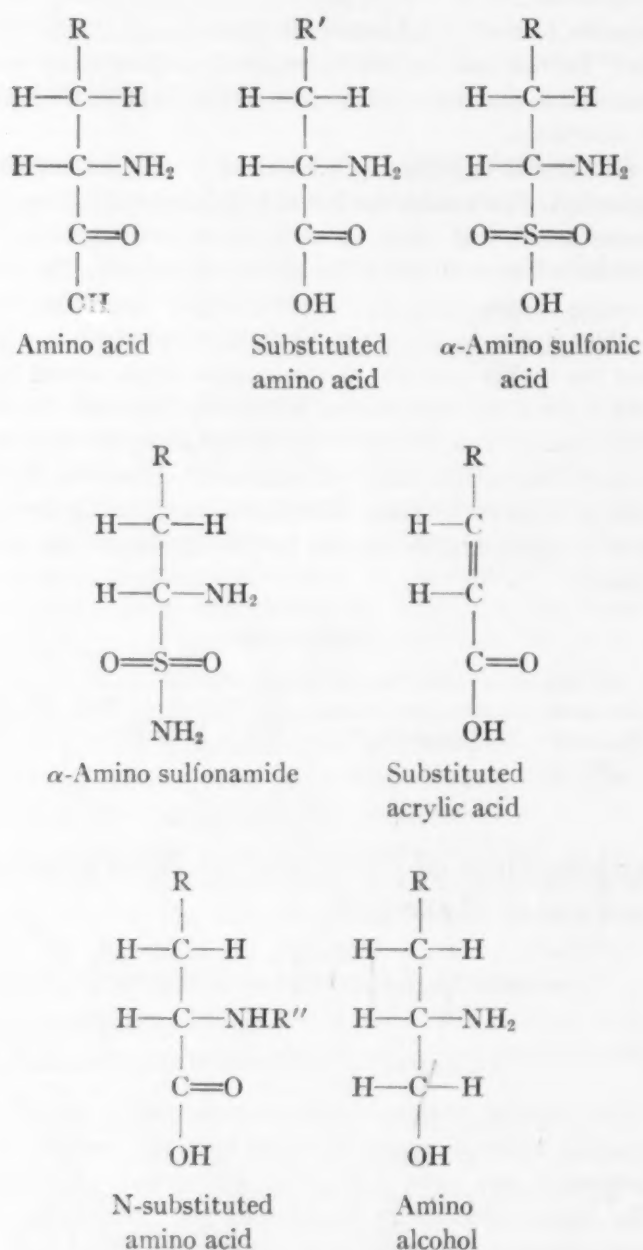
DAVID M. GREENBERG and MARTIN P. SCHULMAN

*Division of Biochemistry,  
University of California Medical School, Berkeley*

The concept that a compound structurally related to an essential metabolite may interfere with the function of that metabolite has been attracting widespread attention (5). The initial stimulus in establishing the "metabolite antagonist" concept has come from the work of Wood and Fildes (6), who demonstrated the antagonistic effect of p-aminobenzoic acid on the action of sulfanilamide. Subsequent investigations from other laboratories (see Roblin, 5) have attempted to find synthetic substances which, as in the case of sulfanilamide, would be incapable of duplicating the physiological action of the metabolite, but would possess great affinity for the same enzyme system and/or other cell constituent with which the metabolite reacts. These attempts have been successful in some instances. In view of the more rapid growth of tumor tissue as compared with normal tissue, it seems plausible to us that it may be possible to interfere with the growth of the malignant tissue to a greater degree than with normal tissue by making use of an appropriate

metabolite antagonist. Thus, regression of a tumor might be affected. Burk and Winzler, in a recent review article (2), have proposed a somewhat similar approach to cancer studies, utilizing competitive vitamins to produce differential vitamin deficiencies in growing and nongrowing tissues.

Since amino acids serve as building blocks in the formation of proteins of normal as well as neoplastic tissue, and further, since proteins are intimately concerned with the functions of protoplasm, our attention was first focused on potential amino acid antagonists (see Fig. 1). It is the purpose of this



R = alkyl or aryl group; R' = variation of alkyl or aryl group; R'' = group replacing H atom of  $-NH_2$ .

FIG. 1. Potential metabolite antagonists of amino acids.

report to furnish a brief account of preliminary experiments concerning the effects of several amino acid analogues on tumor growth. Future work will deal with other types of potential metabolite antagonists and their application to metabolic studies in normal and cancerous tissues.

Because of the relative ease of preparation of  $\alpha$ -amino-sulfonic acids as contrasted with other potential antagonists,

these were used exclusively in this study. Aminomethane sulfonic acid, the compound resembling glycine, was prepared according to the directions of Raschig and Prahl (4);  $\alpha$ -aminoethane sulfonic acid, the sulfonic acid analogue of alanine, according to Backer and Mulder (1);  $\alpha$ -aminoisobutane and  $\alpha$ -aminoisopentane sulfonic acids, analogues of valine and leucine, respectively, according to McIlwain (3); and  $\alpha$ -amino- $\beta$ -phenylethane sulfonic acid, the analogue of phenylalanine, was synthesized in this laboratory. For simplicity, the sulfonic acid corresponding to the amino acid will be referred to as the S-amino acid. Thus,  $\alpha$ -amino- $\beta$ -phenylethane sulfonic acid is S-phenylalanine, while  $\alpha$ -aminoisobutane sulfonic acid will be called S-valine.

Highly inbred "A" mice of both sexes were used. The diet of the animals consisted of Purina Laboratory Chow plus twice-weekly supplements of lettuce. The tumor tissue, Sarcoma A274,<sup>1</sup> was inoculated subcutaneously into the right groin by means of a trocar. Eight days after implantation, the animals were matched according to tumor size. Then, 36 animals with equal-sized tumors (about 1 cm.) were divided into 6 experimental groups of 6 animals each. One of these groups was a control series for all groups, and these 6 mice received no therapy. Each of the remaining 5 groups consisted of 4 test and 2 control animals, the former receiving daily intravenous injections of the S-amino acid while the latter were injected with the equivalent weight of the corresponding dl-amino acid. The groups were: (I) S-glycine, (II) S-alanine, (III) S-phenylalanine, (IV) S-leucine, and (V) S-valine. In all cases the compounds were administered in a total volume of 0.2 ml., the pH of each solution previously being adjusted to 7.4 with 0.2 N sodium hydroxide. Preliminary trials for toxicity level were carried out on other animals, and in all cases daily intravenous doses several times those employed here were well tolerated and no toxic symptoms were observed. After one week, intravenous administration was abandoned, and subcutaneous injections (0.2 ml.), twice daily, were employed. The injections were made at a site far removed from the region of the tumor implantation, namely, into the midscapular region.

The final growth of the sarcoma was not affected appreciably by the administration of the S-amino acid or even by the corresponding dl-amino acid. The experiment, which was terminated 19 days after tumor implantation because several test and control animals succumbed, is being repeated with variations of the experimental conditions and with additional analogues.

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<sup>1</sup> This sarcoma originated at the Wistar Institute. It is a vigorous tissue, palpable three days after implantation and causing death in about three weeks. Sarcoma A274 has been maintained in "A" mice for many generations, 146 transplants having been made since the tumor first appeared. Spontaneous regressions have not been observed to date.



## A Slide Rule for Determining Chicken Red Cell Agglutination Titer

M. C. DAVIES

Lederle Laboratories Division,  
American Cyanamid Company,  
Pearl River, New York

Because of the extensive use of the turbidimetric chicken red cell agglutination (CCA) test at this laboratory in checking production and research on influenza virus, it became desirable to provide a rapid method for evaluating the results. This is customarily done by means of a nomograph<sup>1</sup> with a scale of colorimeter readings along each vertical edge (the nomograph and the slide rule are based on readings obtained from the Klett-Summerson colorimeter). After determining the two virus dilutions which give colorimeter readings bracketing the 50 per cent end-point control, a straight edge is applied to these two readings on the nomograph, and an interpolating factor is read from the horizontal scale above the intersection of the straight edge and the line representing the colorimeter reading of the

ing of standard dilution of unknown which is the higher of the two dilutions bracketing the control value.

The slide rule, which provides a means for determining the fraction  $\frac{Y - X}{Z - X}$  and calibrates this value directly against the

CCA titer, is constructed as shown in Fig. 1. A and B are matching linear scales; C and D are matching logarithmic scales; and the lower four lines are actual CCA values.

Use of the rule requires the following steps:

(1) The lower colorimeter reading (X, above) is taken on the A scale and placed opposite the higher colorimeter reading (Z) on the B scale. This gives a reading on the B scale opposite the A scale index.

(2) The hair line is then set on the D scale at the value obtained from step 1.

(3) The lower colorimeter reading (X) is taken on the A scale and placed opposite the control colorimeter reading (Y) on the B scale. This again gives a reading on the B scale opposite the A scale index.

(4) The sliding portion of the rule is then moved so that the

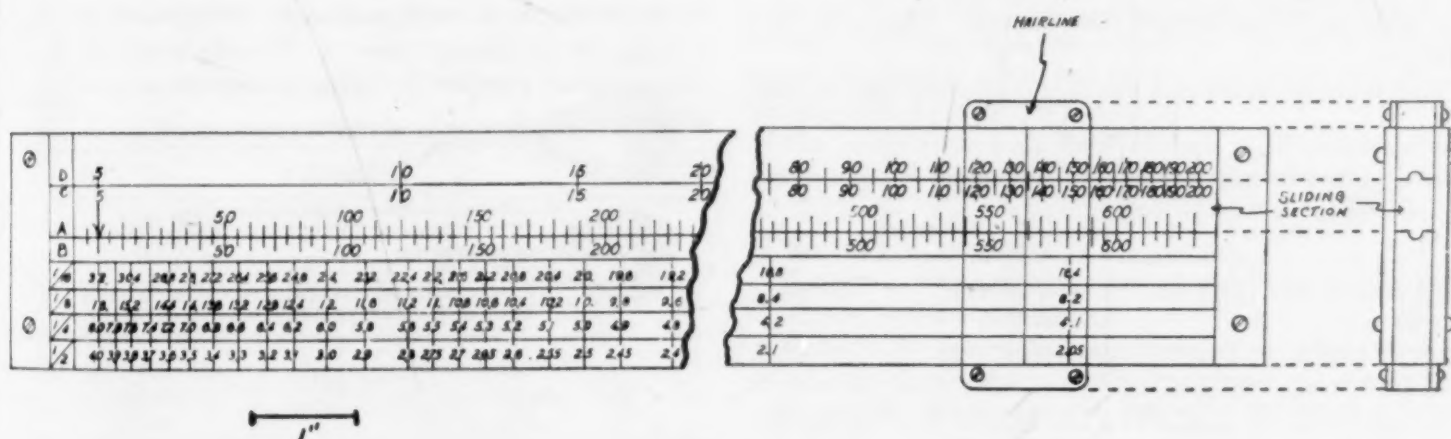


FIG. 1

control. This interpolating factor is then referred to a table, and the desired CCA titer is read therefrom.

A slide rule has been devised in order to carry out these operations more rapidly. The equation involved (1) is

$$\log D = \log S + \frac{Y - X}{Z - X} \log 2,$$

where D is the dilution giving 50 per cent agglutination; S, standard dilution of the unknown which is the lower of the two dilutions bracketing the control value; Y, colorimeter reading of control; X, colorimeter reading of S; and Z, colorimeter read-

value obtained in step 3, taken on the C scale, is under the hair line.

(5) The index on the left end of the sliding portion then is directly opposite the required CCA value on the four bottom scales, which give a range from 1:2 to 1:32, as shown. If desired, this can be extended by adding more lines. The CCA value is read from the appropriate scale; i.e. if the needed figure falls between the 1:2 and 1:4 dilutions, the reading is made on the 1:2 line, etc.

Steps 1 and 2 are subtractions which are usually very simple, so that by performing them mentally further time can be saved.

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<sup>1</sup> NIH directions for the titration of the chicken red cell agglutination (CCA) activity of influenza virus and vaccines according to Hirst's method as modified for use in the Laboratories of the Rockefeller Institute at Princeton, New Jersey.

# Olive Oil as a Solvent for Certain Organic Vapors

R. MACY and B. GEHAUF

Chemical Corps Technical Command,  
Edgewood Arsenal, Maryland

According to Henry's law the solubility of a gas (or vapor) in a liquid is proportional to its partial pressure above the solution, in accordance with the equation  $p = KC$ , where  $p$  is partial pressure of the vapor;  $C$ , its concentration in solution at equilibrium; and  $K$ , a constant. In certain toxicity studies at Edgewood Arsenal, olive oil was used as representative of the body fats, and in connection with proposed calculations of a theoretical nature it was desired to determine to what extent the solubility of the vapors of certain organic compounds in olive oil proceeds in accordance with Henry's law.

The organic compound (about 40 cc.) was contained in a bead bubbler with a diameter of about 40 mm. and capacity of about 80 cc. of liquid, which gave within 2 per cent of saturation at a moderately slow rate of air flow of about 50 cc./minute. Dry air was passed through the bubbler at a measured rate of flow and mixed with a second metered stream of dry air. The diluted vapor was then bubbled through olive oil of known weight, contained in parallel absorption tubes (for check results). The apparatus was all glass except for a few rubber connections, and except for the flowmeters was immersed in a water bath maintained at constant temperature. The concentration of vapor in the air stream was calculated from the flow rates. The saturation concentration in air, shown in Table 1, was based on the assumption that the vapor follows the ideal gas law.

Pompeian brand olive oil was used from newly opened cans.

TABLE 1  
ABSORPTION OF VAPORS\* BY OLIVE OIL AT 20° C.

Nitromethane		Chloroform	
Vapor pressure..... 28.4 mm.		Vapor pressure..... 160 mm.	
Saturation concentration..... 94.9 mg./l.		Saturation concentration..... 1,046 mg./l.	
Saturation in air (%)	Mole fraction in oil†	Saturation in air (%)	Mole fraction in oil†
10.9	0.11	13.2	0.50
17.1	0.16	23.8	0.63
23.8	0.21	43.0	0.75
33.7	0.28		
42.8	0.32		

\* In one experiment with menthol (v.p. = 0.021 mm. and saturation concentration = 0.18 mg./l.) it required 66 hours to reach equilibrium; the mole fraction in oil was 0.19 at 31.8 per cent of saturation in air.

† Molecular weight of olive oil taken as 885. Saponification value = 193.

This oil was brought to constant weight by passing dry air through it for 4 days at 20° C., at the end of which time it was found not to lose weight appreciably. In the solubility determinations (Table 1) at definite partial pressure of vapor it was found that equilibrium was reached in 11-14 hours, as determined by constancy of weight of the absorption tubes.

The solubility data for the two compounds in olive oil are plotted in Fig. 1 in such a way as to compare the experimental data with the ideal solution behavior (the broken line in Fig. 1)

expressed by Raoult's law,  $p = p_0x$ . Since the data represent systems of two miscible liquids, no distinction need be drawn between solvent and solute. When plotted as indicated, chloroform, for example, would conventionally be considered as the

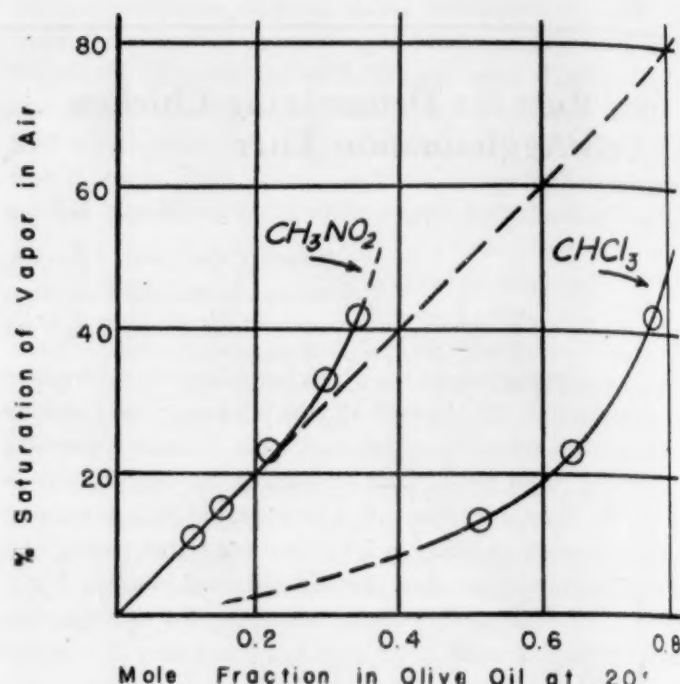


FIG. 1. Deviations from Raoult's law (olive oil as solute); deviations from Henry's law (olive oil as solvent).

solvent and olive oil as solute, with  $p_0$  as the saturation pressure of chloroform;  $x$ , its mole fraction in the solution; and  $p$ , its pressure at the concentration  $x$ . The oil-chloroform system shows a great negative deviation (above-normal solubility of chloroform).

TABLE 2

Concentration of nitromethane (grams/100 grams solution)	Temperature (°C.)
Solubility of nitromethane in olive oil	
9.31	3.0
10.79	17.1
11.94	24.5
14.65	41.2
Solubility of nitromethane in water	
9.32	0.7
10.39	15.8
11.29	25.7
13.17	40.5

The oil-nitromethane system, considering nitromethane as solvent, follows Raoult's law fairly well up to a concentration of about 0.2 mole fraction of nitromethane. At higher concentrations than this there is a pronounced positive deviation (below-normal solubility of nitromethane). Although the vapor solubility work was not carried out at higher concentrations than those shown in Fig. 1, it was found that nitromethane and olive oil form a two-liquid system at 20° C. when the mole fraction of nitromethane in the oil phase reaches 0.67 (interpolated from data in Table 2).

To interpret the data on the basis of Henry's law, olive oil



is regarded as the solvent, which corresponds with the experimental procedure. Dilute solutions with nitromethane as solute follow this law to the extent that Raoult's law is followed as stated above. As to chloroform as solute, data were not obtained at low enough concentrations to determine if the ratio between concentration in the oil phase and pressure in the vapor phase would be constant in dilute solution.

*Solubility of nitromethane in olive oil and in water.* The solubility determinations were made by the synthetic method, using total quantities of about 5 grams in sealed glass tubes. Temperatures at which turbidities appeared or disappeared could be determined over a range of about 0.5°. The data obtained are recorded in Table 2.

## New Rectal Culture Tube<sup>1</sup>

JOSEPH FELSEN

*The Dysentery Registry, New York City*

The importance of examining material obtained directly from the intestinal wall in the diagnosis of infectious diarrheas has been stressed in recent years by students of the subject. The sigmoidoscopic mucosal crypt aspirator has been described elsewhere (1). It is essentially a heavy-walled capillary tube 40 cm. in length, with the distal 2.5 cm. bent 15 degrees for better approximation of the tip to the intestinal mucosa. At the point of bending, the capillary canal is blown into a small elliptical dilatation of sufficient size to hold two or three drops of aspirated material. This instrument requires the use of a sigmoidoscope, and it is applied to the mucosa by direct vision.



FIG. 1

The new rectal culture tube (Fig. 1) may be used without the aid of the proctoscope or sigmoidoscope. It consists of a metal outer tube 18 cm. in length with a round opening 0.5 cm. in diameter (A) near its distal end. An inner tube (B) screws into a detachable receptacle (C) with smooth tip 1.4 cm. in length and containing a shallow trough. When fitted together, the trough coincides with opening A in the outer tube and may be opened or closed by a quarter turn at D, which represents the proximal end of the inner tube, containing a cotton filter (E), to which a small aspirating rubber bulb (F) is attached.

In actual use, the sterile, assembled, rectal culture tube (without rubber bulb), contained in a stoppered sterile test tube, is removed and lubricated at its distal end (A) by dipping it into sterile glycerine. The rubber bulb is attached to the proximal end. The metal tube, with receptacle at A closed, is then inserted into the rectum for a distance of approximately

10 cm. The inner tube is given a quarter turn, indicated by an arrow at G and a small stop at H. This opens the receptacle at A, which is then applied to the rectal mucosa. The exudate is aspirated into the receptacle, which is promptly closed by a quarter turn in the reverse direction, and the instrument is removed from the bowel. The material may be cultured and wet smear studies made at once by opening the receptacle and blowing out the aspirated material on suitable culture media and slides; or the closed instrument can be dropped into the original test tube and transported to the laboratory. Sterilization and cleaning of the three simple components of the instrument are effected quickly, efficiently, and economically.

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## Comparison of Hematocrit Methods<sup>1</sup>

PAUL L. McLAIN

*Department of Physiology and Pharmacology,  
School of Medicine, University of Pittsburgh*

In connection with a study on relative corpuscle and serum (plasma) volumes in blood, selection of a reliable hematocrit method has become a matter of primary concern. While the physical factors which affect the final volume of centrifugally packed corpuscles are well known (4, 5, 6), this information is frequently ignored in practice, with the result that many different forms of apparatus have been proposed and are in common use for routine work. In these experiments, three such methods were compared, first, as to their reliability in detecting known dilutions of blood, and second, as to differences among the absolute measurements obtained.

The comparisons were made on each of 10 samples of defibrinated beef blood, and on 3 dilutions of each sample with its own serum. The dilutions contained 90, 80, and 50 per cent of whole blood, respectively.

The types of hematocrit tubes employed and the details of centrifugation were as follows:

*Method 1.* Straight-walled glass tubes of the Daland (1) type, 50 mm. long with 1-mm. bore, were rotated in a standard Daland head at 12,000 r.p.m. and 4.7-cm. effective radius (centrifugal force, approximately  $7,500 \times G$ ) until constant sediment volumes were obtained. This usually required less than 10 minutes of actual spinning and in every instance caused translucence of the sediment column (3).

*Method 2.* Straight-walled glass tubes of the Wintrobe (7) type, 11.5 cm. long with 3-mm. bore, sealed at one end, were rotated in an International centrifuge at 3,000 r.p.m. and 14-cm. effective radius (centrifugal force, approximately  $1,400 \times G$ ) for one hour.

*Method 3.* Ordinary, tapered, graduated, 15-ml. centrifuge tubes, containing 10 ml. of blood, were rotated at the same time and under the same conditions as described for Method 2, following closely the recommendations of Haden (2).

All tests were in duplicate, the pairs being centrifuged simultaneously. Paired observations differed by 1.0 volume per cent or less in 95 per cent of the tests.

<sup>1</sup>The author wishes to acknowledge the assistance given him by Clay-Adams Company, 44 East 23rd Street, New York City, in the designing of this tube.

<sup>1</sup>Presented before the 31st Annual Meeting of the Federation of American Societies for Experimental Biology, Chicago, Illinois, May 18-22, 1947.

Typical results are presented in Table 1. Table 2 summarizes the data, giving the major statistical constants for relative volumes of red cells, errors in prediction of dilution, and deviations from the results of an arbitrary reference standard.

TABLE 1

Sample No.	Whole blood in mixture (%)	Relative volumes of red cells (Vol. %)		
		Method 1	Method 2	Method 3
4	100.0	40.5	41.1	41.3
	90.0	35.5	36.8	37.5
	80.0	32.0	32.4	34.1
	50.0	20.0	19.8	20.4
5	100.0	44.5	47.0	48.3
	90.0	40.5	42.0	43.0
	80.0	36.0	36.6	37.2
	50.0	22.5	22.9	22.6
6	100.0	46.0	46.2	47.8
	90.0	41.0	41.9	42.3
	80.0	36.5	36.8	39.0
	50.0	23.0	23.3	24.3

With regard to the actual hematocrit values obtained, the methods yielded slightly different but statistically indistinguishable means and variance for the entire series and at each dilution level (Table 2, A).

TABLE 2

Method No.	Obs.	Mean	S.E.	Max.	Min.	Alg. mean	Distribution of errors		
							Under 1%	Under 3%	Under 5%
A. Relative volumes of red cells									
	No.	Vol. %	Vol. %	Vol. %	Vol. %				
1	44	35.5	1.3	52.0	18.5				
2	38	35.7	1.6	53.5	18.3				
3	38	36.6	1.6	54.1	19.0				
B. Errors in prediction of dilution									
	No.	%	%	%	%	%	% of obs.	% of obs.	% of obs.
1	33	0.95	0.12	2.78	0.00	-0.29	51.5	100.0	100.0
2	28	2.05	0.29	5.50	0.00	-1.89	42.9	75.0	96.4
3	28	1.95	0.28	6.40	0.00	-1.34	32.1	78.5	96.4
C. Deviations from results by Method 1*									
2	38	2.20	0.26	7.30	0.33	+1.51	13.2	73.7	92.1
3	38	3.53	0.31	8.54	0.00	+3.53	10.5	39.5	81.5

\* Method 1 was arbitrarily taken as a standard to illustrate the magnitude of differences among the methods.

With respect to quantitative detection of blood dilution, the various methods yielded the errors summarized in Table 2, B. The mean error for Method 1 was significantly lower than those for the remaining methods. Further, all errors for Method 1 were less than 3.0 per cent of the expected value, an accuracy attained in about three-fourths of the tests for Methods 2 and 3. The algebraic mean for all methods was negative in sign, indicating a preponderance of relatively low readings for the di-

luted samples. This feature was much less prominent for Method 1 than for the other methods. Methods 2 and 3 were about equivalent in reliability, while Method 1 was definitely superior from the standpoint of revealing degree of blood dilution.

Table 2, C, contains an analysis of individual deviations among the methods, based on Method 1 as an arbitrary reference standard. The difference between the means was significant, and the variance indicated definite disparity between the two methods, although the total range was about the same for both. With regard to distribution of these deviations, Method 2 was superior to Method 3, especially as to errors less than 5.0 per cent. The algebraic mean for both methods was positive in sign, indicating a tendency for both of these procedures to yield high results in comparison with Method 1. Results for Method 3 exceeded those for Method 1 in 95 per cent of the observations. Readings for Method 3 exceeded those for Method 2 in about three-fourths of individual comparisons, while the latter were greater than the values for Method 1 in roughly the same proportion. Degree of dilution did not significantly affect the relative differences between the values for Method 1 and those for the other methods.

The three hematocrit methods studied, involving widely different varieties of tubes and different conditions of rotation, produced results which, on the average, were not seriously divergent. At the same time, differences among the methods appeared in the comparisons of results on individual samples and in the accuracy with which known dilutions of blood were revealed by hematocrit observations. Method 1 was the most satisfactory from both points of view, yielding, in general, the lower hematocrit values and measuring degree of dilution with an average error of less than 1.0 per cent. The further advantages of speed and minimal blood requirement were obvious. Method 2 was generally more satisfactory than Method 3. Although it was no more accurate in revealing dilution, it gave results more nearly approximating the values for Method 1, and the distribution of its errors and deviations was more favorable. The amount of blood required was not excessive, and no special equipment, aside from the tubes, was necessary. Method 3 was considered the least reliable. Its accuracy in detection of dilution, while comparing favorably with Method 2, was distinctly inferior to Method 1. Furthermore, results obtained by Method 3 were consistently higher than those of other methods—a significant point in view of the general criticism of centrifugal methods, namely, that cell volumes so determined are always high because of the presence of fluid in the sediment.

The study indicated that, for problems demanding a high degree of accuracy, commonly employed hematocrit techniques are not equivalent, and that average comparative results are not necessarily trustworthy criteria of the general reliability of such methods.

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# The Halogen-Metal Interconversion Reaction and Its Application to the Synthesis of Nicotinic Acid Labeled With Isotopic Carbon<sup>1</sup>

ARTHUR MURRAY III, W. W. FOREMAN,  
and WRIGHT LANGHAM

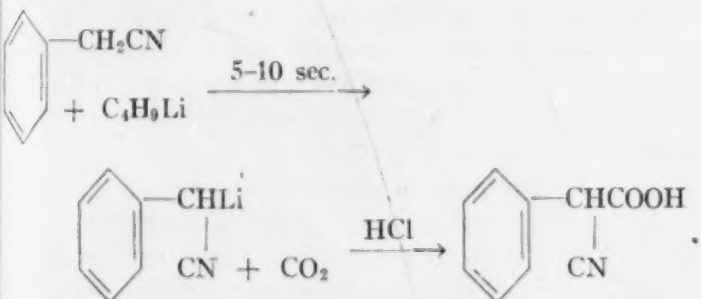
University of California Los Alamos Scientific Laboratory,  
Santa Fe, New Mexico

The halogen-metal interconversion reaction is essentially a double decomposition reaction between an organoalkali compound (usually a metal alkyl) and an aryl halide:

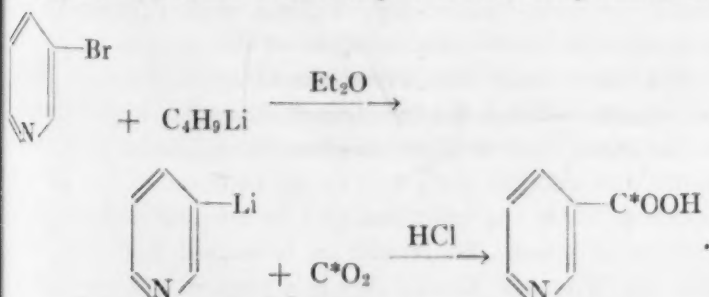


Gilman, Langham, and Moore (1) made an extensive study of the application of the above reaction to the preparation of a number of unusual organolithium compounds. The extremely short reaction times required permit the preparation of organometallic compounds having additional functional or reactive groups, a feat not possible in the usual Grignard preparation. Like the Grignard reaction, however, these reactions yield carboxylic acids when carbonated with CO<sub>2</sub>.

The extreme rapidity of the exchange reaction is not confined to the halogen-metal interconversion alone but may be evidenced in special cases of the hydrogen-metal exchange. For example, if benzyl cyanide is treated with n-butyl lithium, a 55 per cent yield of phenylcyanoacetic acid results from carbonation within 5-10 seconds. The reaction proceeds according to the following equation:



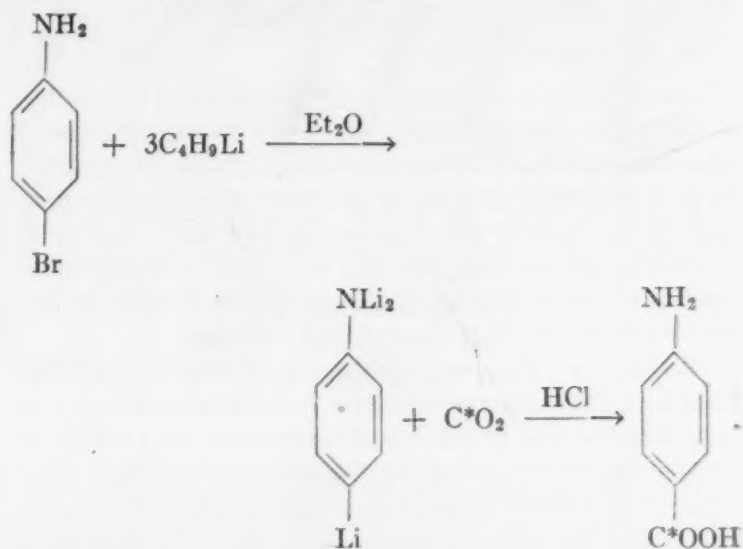
Using n-butyl lithium and 3-bromopyridine according to the method reported by Gilman and Spatz (2), 3-pyridyl lithium was prepared. The reaction was scaled down to the millimol level and carbonation effected with C<sup>13</sup>O<sub>2</sub> and C<sup>14</sup>O<sub>2</sub>, generated by treating the respective isotopic barium carbonates with concentrated H<sub>2</sub>SO<sub>4</sub>. Isotope-labeled nicotinic acid (3-pyridine carboxylic acid) resulted according to the following reactions:



A reaction employing 21 millimols of BaC<sup>13</sup>O<sub>3</sub> resulted in an 82 per cent crude yield of nicotinic acid having a C<sup>13</sup> content of

0.9 isotopic per cent. When the reaction was repeated using 0.8 millimol of BaC<sup>14</sup>O<sub>3</sub>, the crude yield of C<sup>14</sup>-labeled nicotinic acid was 62 per cent. The material had a specific activity of approximately 8 microcuries/mg.

The interconversion reaction between n-butyl lithium and p-bromoaniline (3) is being employed for the synthesis of p-aminobenzoic acid labeled with isotopic carbon according to the following reactions:



A reaction using 3 millimols of ordinary BaCO<sub>3</sub> has resulted in a 21 per cent yield of p-aminobenzoic acid of high purity.

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## Analytical Determination of Basic Groups in Amino Acids and Proteins

D. P. GRETTE

Research Laboratories, Swift & Company, Chicago

In the course of work upon proteins, it has been found that the basic groups of amino acids (except arginine and histidine) or of proteins can be determined rapidly and simply.

The sample (0.2 gram of amino acid or 2 grams of protein) is dissolved at room temperature in 100 ml. of 0.05 N Ca(NO<sub>3</sub>)<sub>2</sub> solution saturated with Ca(OH)<sub>2</sub>. An excess of solid Ca(OH)<sub>2</sub> (0.5 gram) is added, and the mixture is shaken repeatedly during two hours. It is then filtered and protected from the air to avoid carbonation. An aliquot is titrated with standard HCl (0.05 N). A blank containing no amino acid or protein is prepared at the same time and carried through simultaneously with the sample. The difference between sample and blank represents the basic groups in the sample and may be expressed as the per cent of basic nitrogen in the sample. Results on pure amino acids are accurate to within 1.5 per cent error.

This method has some advantages over the Sorensen method, particularly with regard to proline and hydroxyproline, which can be titrated accurately by the present method.

A similarly simple method of determining the acid groups of amino acids and proteins has been sought, but completely satisfactory results have not been obtained. Use of picric acid has given the best results to date.

<sup>1</sup> This paper is based on work performed under Contract W-7405-Eng-36 with the Manhattan Project at the Los Alamos Scientific Laboratory of the University of California.

# Book Reviews

**Fundamental chemistry: an elementary textbook for college classes.** (2nd ed.) Horace G. Deming. New York: John Wiley; London: Chapman & Hall, 1947. Pp. xvi + 745. (Illustrated.) \$4.00.

Teachers who are interested in a presentation of elementary chemistry which encourages thought and the development of a questioning and scientific attitude on the part of the student, teachers who are desirous of a treatment which is not repetitious to the student who has already had some training in chemistry, and students who are concerned with improving their own basic knowledge of chemistry will all welcome this new edition of Deming's *Fundamental chemistry*.

This book is truly a worth-while one and may be read with profit by teacher and student alike. It is logically written in a prose which is easy to follow and understand. It is orderly in its presentation and accurate in its information. It is thoroughly modern in its thesis, its approach, and its treatment of theory and fact. It is designed to present chemistry as a "manner of thinking" rather than as a "collection of facts," and to this end it encourages the student to thought through well-chosen questions, problems, diagrams, and illustrative examples.

Although addressed primarily to the chemistry major and to the individual who has some previous knowledge of the subject, the book can be adapted to others by limiting assignments to indicated selected topics. Theoretical concepts are presented gradually in conjunction with factual material, although treatment of the latter is much less comprehensive than in the ordinary beginning textbook, particularly with regard to the metallic elements. The book contains much explanatory information beyond that contained in most elementary treatments and is to be commended for its stress upon rigid definitions and its dimensional and reasoning approach to problems.

The book is free from significant errors.

THERALD MOELLER

University of Illinois, Urbana

**Visible speech.** Ralph K. Potter, George A. Kopp, and Harriet C. Green. New York: D. Van Nostrand, 1947. Pp. xvi + 441. (Illustrated.) \$4.75.

This book is a record of the work of the Bell Telephone Laboratories in converting speech sounds instantaneously and continuously into readable visible patterns.

The first section of the book briefly and nontechnically describes the "Direct Translator," which produces the pattern from the sound, and states that a group practiced with the instrument is able to communicate vocally among its members totally without benefit of hearing.

The second section comprises 225 pages of superbly illustrated and illuminated lessons in the art. More than 500 cuts illustrate "vocabulary" and tie the patterns to the vocal mechanisms which produce the sounds.

A third section (154 pp.) considers the principles of Visible Speech in relationship to such fields as speech correction and indicates its use in teaching vocal speech to the totally deaf.

Some space is also taken up with cuts of the singing voice and even of instrumental music.

The book will be of interest to every student of speech and speech correction. It illuminates the mechanics of vocal sound production exceptionally well and completely.

Two of its authors are practiced speech teachers; the third, an engineer. In style it is nontechnical and exceedingly readable.

T. E. LYNCH

The Brush Development Company, Cleveland, Ohio

**Glacial geology and the Pleistocene epoch.** Richard Foster Flint. New York: John Wiley; London: Chapman & Hall, 1947. Pp. xviii + 589. (Illustrated.) \$6.00.

According to the Preface, this book presents the principal facts of glacial geology and the Pleistocene epoch in compact form for geologists, ecologists, archaeologists, geographers, soil scientists, and other students of the Pleistocene. It is a summary of established data for the entire globe, and it indicates areas in which research is greatly needed.

The 23 chapters may be grouped as follows: (1) Summary; (2) The nature and properties of existing glaciers; (3) The nature of the record left by previous glaciers of the Pleistocene; (4) Glaciation of North America; (5) Glaciation of Europe; (6) Glaciation outside North America and Europe; (7) Relations between glaciation, mountain uplifts, changes in level of land and sea, former great lakes in dry continental areas, and the migrations of animals and plants; and (8) The relation of climatic fluctuations to glaciation.

This simplified table of contents does not give any idea of the wealth of detail in the book. Approximately 1,000 titles, of which about 17 per cent are in languages other than English, are listed in the bibliography. There are 30 tables, 20 photographs, 30 diagrams, and 40 maps, of which 6 are folded plates at the back of the book. The reader will wish to refer, in addition, to the large Glacial Map of North America (Flint, *et al.*, 1945), published separately by the Geological Society of America.

The author goes into considerable detail concerning a definition of the term Pleistocene, and his suggestion that the terms Tertiary, Quaternary, Recent, and Postglacial be dropped from formal nomenclature.

Yet this is more than a reference book. It is written with enthusiasm and makes interesting reading. One feels oriented at the same time to the achievements of glacial geologists during the last 100 years and to the prospect of further research to fill in the enormous gaps in our knowledge of the Pleistocene epoch. The reader is impressed with the fact that the Wisconsin Glacial Age is a present and continuing reality.

It is a satisfaction to note that the book is dedicated to the memory of the young glaciologist, Max Demorest (1910-1942).

FRANCIS D. HOLE

Soils Building, University of Wisconsin